SCIENTIFIC OPINION

Scientific Opinion on the safety and efficacy of using recycled hot water as a decontamination technique for meat carcasses[1]

EFSA Scientific Panel on Biological Hazards (BIOHAZ)[2,4]

EFSA Panel on Contaminants in the Food Chain (CONTAM)[3,4]

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ABSTRACT

For carcass decontamination purposes, only use of potable water is currently allowed in the EU. However, recycling (i.e. reusing after reheating) of the water used for carcass decontamination has been practiced in some countries (e.g. Canada, Denmark), because environmental and energy-preserving reasons. In this document, potential microbiological and abiotic risks for carcasses associated with recycled hot water decontamination, and related control options, were considered. It has been concluded that the decontamination efficacy of recycled hot water does not differ significantly from that of hot potable water. With recycled hot water, only microbiological risks associated with heat-resistant bacterial spores (*C. botulinum, C. perfringens, C. difficile* and *B. cereus)* are relevant. These risks can be controlled through ensuring that recycled hot water is verifiably subjected to such reheating and frequency of renewal regimes which ensure that the microbiological risk in recycled water is not higher than in hot potable water. For abiotic risks, the only concern with recycled hot water derives from the potential presence and accumulation of residues of veterinary drugs and other chemical contaminants not addressed in Council Directive 98/83/EC in the water for decontamination of poultry carcasses.

KEY WORDS

Contaminants, decontamination, HACCP, recycled hot water, spores, veterinary medicinal products

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SUMMARY

Following a request European Commission, the Panel on Biological Hazards (BIOHAZ) was asked to deliver a scientific opinion on safety and efficacy of using recycled hot water as a decontamination technique for meat carcasses. The Panel on Contaminants in the Food Chain (CONTAM Panel) was asked to assess the abiotic risks.

In particular the BIOHAZ Panel was asked to: assess the efficacy of this decontamination technique in terms of reduction of surface contamination; evaluate the abiotic and microbiological risks for the carcasses arising from the carcasses and/or the water system when using recycled hot water and suggest control options; identify and define criteria for the HACCP in order to obtain the expected efficacy and to control the possible risk, e.g. the required range of temperature of the recycling water, criteria as fat modifications, denaturation of protein, the frequency of the renewal of the water. According to the remit of this Opinion, the microbiological risks considered only relate to bacteriological and protozoan pathogens, whilst prions are not dealt with.

The published available data on the efficacy of recycled hot water decontamination are very limited and relate only to treatments of bovine and porcine carcasses and only to spray and deluge[5] application techniques. Nevertheless, the available data have shown no significant differences in decontamination efficacy, in terms of microbial reductions achievable on carcasses, between hot potable and hot recycled water.

It was concluded that the application of proper heating regime of recycled water is the main option to control vegetative bacterial cells and protozoan parasites, whereas microbial toxins are not significantly produced and/or are inactivated in the process.

The microbiological risks in the recycled water of main potential concern derive from heat-resistant bacterial spores[5] such as *C. botulinum, C. perfringens, C. difficile* and *B. cereus*, however there is a lack of data on the extent of carcass contamination with spores, their germination and inactivation during the recycling process, and the potential for accumulation, during the operations. In order to control the risk related to spores, the BIOHAZ Panel concluded that the best option is to define a proper criteria for the HACCP in order to ensure that the microbiological risk in recycled water is not higher than in hot potable water.

Concerning the abiotic risks, the CONTAM Panel concluded that, if compliance of recycled hot water with the existing chemical criteria for potable water is ensured, it is unlikely that there would be an increased abiotic risk using recycled hot water for decontamination of carcasses as compared to hot potable water decontamination treatment. However, the existing criteria for potable water neither include all chemical contaminants nor veterinary medicinal products, which might contaminate recycled hot water.

The criteria for the HACCP in order to obtain the expected efficacy and control the possible risk include the minimal heating temperature/time regime for, and frequency of renewal of, recycled water before its application on carcasses. This must ensure compliance with existing microbiological criteria for potable water, and prevent accumulation of heat resistant spores. Different time/temperature heating regimes and frequency of renewal of water can be used but should be microbiologically validated, continuously monitored by instrumental measurements, verified periodically by microbiological testing of water, and documented. It is concluded also that the compliance with the chemical criteria for potable water need to be verified for recycled hot water by periodic chemical analysis of the water and documented. For recycled hot water applied on carcasses, different temperatures and application techniques-related parameters can be used, but their critical values have to be specified, validated, monitored, verified and documented in the same way as with hot potable

5 See Glossary

water decontamination. Furthermore, the absence of residues of veterinary medicinal products in the recycled hot water used for decontamination of poultry carcasses has to be verified by periodical testing, and documented.

It is recommended that further research should be encouraged on the presence and potential accumulation of bacterial spores in the water for decontamination of carcasses of all animal species, and on the potential presence and accumulation of residues of veterinary drugs and other chemical contaminants in the hot recycled water for decontamination of poultry carcasses.

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BACKGROUND AS PROVIDED BY EUROPEAN COMMISSION

Regulations (EC) No 852/2004 and (EC) No 853/2004 only allow the use of potable water on carcases. Potable water is defined in Council Directive 98/83/EC on the quality of water intended for human consumption, in any of its physical forms.

Some Third Countries and Members States have an increasing interest in using alternative methods, such as hot water as decontamination technique.

The Commission considers that the use of hot potable water to remove surface contamination on carcases may be allowed provided that:

- the hot water used is potable, as laid down in Article 3(2) of Regulation (EC) No 853/2004; and in Council Directive 98/83/EC;
- the hot water application does not result in any irreversible discolouration of the meat; and
- all the relevant requirements of Regulations (EC) No 852/2004, (EC) No 853/2004 and (EC) No 854/2004 for the production of fresh meat are respected. The Scientific Committee on Veterinary Measures relating to Public Health in its opinion on 14 and 15 April 2003 on "the evaluation of antimicrobial treatments for poultry carcases" concluded that decontamination can constitute a useful element in further reducing the number of pathogens provided that an integrated control strategy is applied throughout the entire food chain, including hygienic measures applied at primary production, during transport and in the slaughter and processing plant.
- with regard to Article 5 of Regulation (EC) No 852/2004, when steam or hot potable water decontamination is used, it shall be considered as a potential Critical Control Point in the HACCP-based approach.

However, the use of the technique of recycling hot water in a closed system to remove contamination on carcases could represent a possible risk. Therefore the Commission, in consultation with the Member States, decided that further scientific advice was needed.

Canada and Denmark have provided reports on this topic which could provide background information for this assessment.

TERMS OF REFERENCE AS PROVIDED BY EUROPEAN COMMISSION

EFSA is asked to issue a scientific opinion on the safety and efficacy of using recycled hot water as a decontamination technique for different species covered by definitions of Regulation No 853/2004 and in particular:

1. To assess the efficacy of this decontamination technique in terms of removal/reduction of surface contamination.

2. To evaluate the abiotic[6] and microbiological risks for the carcases arising from the carcases and/or the water system when using recycled hot water and suggest control options.

3. To identify and define criteria for the HACCP[4] in order to obtain the expected efficacy and to control the possible risk:

    - The required range of temperature of the recycling water;
    - Other criteria which should be considered (e.g. fat modifications, denaturation of proteins);
    - The frequency of the renewal of the water and/or suggested parameters to verify this.

6 See Glossary

ASSESSMENT

1. Introduction

Modern microbial food (including meat) safety assurance is based on a farm-to-fork principle that involves a wide range of coordinated control measures applied at all relevant steps in the food chain (EFSA, 2009). The main, fundamental approach to these measures is of preventative nature and relies on good hygienic practices so as to completely avoid or at least minimise microbial contamination. It has been recognised that in some food industry processes total prevention of microbial contamination is unachievable solely through hygienic measures and certain microbial contamination is unavoidable. Such processes especially include abattoir operations, i.e. slaughter and dressing of animals.

Carcass contamination can be directly and/or indirectly linked with the animal faeces potentially carrying most relevant microbial foodborne pathogens such as *Campylobacter*, *Salmonella* and verotoxigenic *Escherichia coli*. For these reasons, even if the incidence/levels of such "unavoidable" contamination are relatively low, the associated risk for public health can be considered as unacceptable. This is because often a connection exists between the foodborne pathogens' status of carcasses and the risk of related illnesses. For example, the WHO/FAO risk characterisation of the salmonellae in broilers stated that the relationship between the carcass prevalence of *Salmonella* spp. and the attributable risk of human salmonellosis is essentially linear (WHO/FAO, 2002).

As a part of good hygienic/manufacturing practices, carcasses of all slaughter animals are routinely washed with cold potable water at the final point of the slaughter line, or at two or more points, e.g. in case of pigs and, particularly, poultry carcasses. However, it is known that while cold-water washing may reduce numbers of bacteria on carcasses when numbers are relatively high, it is less effective when they are relatively low (Gill and Badoni, 2003). Generally, even when carcass washing is applied in the most effective way, it can remove only up to 1 log unit of the meat microflora (Varnam and Sutherland, 1995; Sheridan, 2004). Furthermore, it is questionable whether those cold-water washing reductions are real, or whether the microflora is partly just redistributed from heavier contaminated sites over the carcass onto less contaminated ones leading to a false interpretation. Several carcass washes with water are regularly used in poultry abattoirs; still, overall relatively modest reduction of surface contamination by 1 to 2 log units is achievable (SCVPH, 1998). Therefore, microbial reductions achievable only by cold-water washing are widely considered to be of no practical significance (Sheridan, 2004).

To additionally reduce human health risk from carcass microbial contamination, i.e. beyond the reductions achievable by good hygienic practices, a complementary approach of employing various treatments aimed at the elimination of microorganisms from carcasses can be considered within the regulatory frame. It is important to note that science-based risk management policy considers that food (including carcass) treatments to reduce microbial surface contamination should only be considered as an additional measure, providing extra assurance for situations of accidental and unknowingly high contamination, following the application of good hygienic/manufacturing practices, without being considered as substitutes for them (SCVPH, 1998; SCVPH, 2003; EFSA, 2006; EFSA 2010). Such complementary interventions of decontamination have to be incorporated into the HACCP system of managing processes for pathogen control.

A large number of different antimicrobial ("decontamination") treatments have been developed, and evaluated, and some applied in certain third countries (Acuff, 2005; Bacon et al., 2000; Dorsa, 1997; Feirtag and Pullen, 2003; Chen et al., 2005; Huffman, 2002; Smulders and Greer, 1998; Sofos, 2005; Sofos and Smith, 1998). It is important to note that, with a given treatment, higher microbial reductions are usually achieved under experimental compared to commercial conditions. The published carcass decontamination techniques are based on a range of physical and chemical treatments. Based on the knowledge accumulated to date, it is assumed that currently available decontamination treatments, generally, can only reduce the microbial contamination level on, but

cannot completely eliminate it from carcasses. Furthermore, the ultimate effectiveness of antimicrobial treatments can vary markedly. The variability is due to several factors that can be variable themselves, including high dependence on the initial microbial load, temperature, exposure duration, pH, hardness of the water, bacterial attachment to meat, biofilm formation and the presence of fat or organic material in water (SCVPH, 2003). In addition, the intensity of application of carcass decontamination treatments is limited by potential undesirable changes of meat sensory qualities. Such negative effects may be avoided, while antimicrobial activity is optimized through application of lower intensity sequential or simultaneous treatments in the form of multiple interventions or hurdles, aiming at synergistic or additive effects (Bacon et al., 2000; Sofos, 2005; Sofos and Smith, 1998).

Overall, among interventions employed under commercial conditions, treatments of carcasses (e.g. porcine and bovine) using hot water or steam are often advocated as the most effective and reliable (Phebus et al., 1997; Gill and Bryant, 2000), and, hence, are commonly used by meat industry in some countries e.g. USA, Canada, Australia and Denmark. With respect to hot water treatments, repeated use of water collected after carcass treatments (i.e. re-cycling) following re-heating is practiced by some abattoirs in some countries. Recirculating the water offers processors both cost and carbon savings in both energy (less heat is required to reheat the recirculated water, although energy is required to treat and recirculate the water) and water use.

In principle, the use of re-cycled hot water for carcass decontamination could be justifiable and acceptable only if it is not associated with increased meat safety-, public health- and product quality-related risks, and if applied under controlled, verifiable conditions incorporated within the HACCP-based meat safety process management system. Therefore, the main scope of this document is: a) to analyse current knowledge and available data in order to assess the decontamination efficacy (see Glossary) of the re-cycled hot water carcass treatment as well as the associated abiotic and microbiological risks; and b) to identify conditions/parameters required for risk control. According to the ToRs, in this Opinion, the microbiological risks considered only relate to bacteriological and protozoan pathogens, whilst prions are not dealt with. Where relevant, these considerations include comparisons between carcass treatment with recycled hot water and hot potable[7] water, as well as no treatment option (cold water washing treatment is applied to all the carcasses). Because a number of recycled hot water treatments differing in respect to some or many technical characteristics affecting their effectiveness probably exist, they cannot be and are not evaluated individually here; rather the considerations are conducted in a generic, more universally applicable way.

The CONTAM Panel of EFSA was responsible to take care of the evaluation of the abiotic risks. On the other hand, in accordance with the remit of the BIOHAZ Panel, environmental protection-related issues are not dealt within this document.

7 The microbiological and chemical parameters of potable water to which the opinion refer to, are those listed in the Part A and B of the Directive 98/83/EC about the quality of water intended for human consumption. The indicator parameters listed in the Part C refer to consumers subjectivity, therefore are out of the scope of this opinion.

2. Systems and techniques used to apply hot water decontamination treatments

2.1. Hot water decontamination systems

Hot water may be applied for meat decontamination by spraying at higher, or low, pressures, deluging with cascading sheets of hot water, or by immersion (dipping) of the product (Corry et al., 1995; Sofos and Smith, 1998; Dincer and Baysal, 2004; Hugas and Tsigarida, 2008). According to Sofos and Smith (1998), each procedure has advantages and disadvantages. Immersion may be more applicable to smaller animals (e.g., poultry) or meat cuts, while spraying at high pressures may remove visible soil but not achieve the desired high temperatures and may generate aerosols. Low pressure rinsing yields higher tissue temperatures, while deluging with hot water achieves higher temperatures throughout irregularly shaped carcasses or cuts. Hot water treatments may take place at pre-evisceration and after final washing during slaughter and dressing, and after chilling during carcass deboning on meat cuts and trimmings for manufacturing ground beef (Sofos and Smith, 1998).

2.2. Spraying

Extensive studies have been carried out on carcass spray treatment in the UK by Bailey (1971), in Ireland by Kelly (1981, 1982), in the USA by Anderson and co-workers on the CAPER system (Anderson et al., 1987), and in Australia by Smith and Davey (Davey, 1988; Davey and Smith, 1989). Together, these studies provide essential information on the parameters that affect the effectiveness of spray treatments. Physical parameters include spray pressure and flow rate, and nozzle type, configuration and the angle of spray. In addition, other variables such as tissue type, or temperature of treatment also affect the efficacy of decontamination procedures. Many of these have been discussed in Pordesimo et al. (2002) review of spray treatment; however this review concentrates mainly on relatively recent US based studies. The parameters reported in this review are shown in the Appendix A. Naturally raising the temperature of the water increases the reduction in microbial counts. However, there are practical problems in using hot water, for instance, a spray-jet water rapidly loses heat by evaporation. Studies have shown that the maximum impact temperature on the carcass from a spray placed 30 cm away and supplied with water at 90°C is approximately 63°C (Bailey, 1971). This problem has lead to the development of various cabinet systems (Graham, 1979).

2.2.1. Manual sprays

Spray treatment was originally introduced in the UK as a method of removing visible dirt from carcasses, following the abolition of the wiping cloth in 1968. Bryce-Jones's (1969) initial trials investigated the effect of spray treatment on beef and lamb carcasses. His conclusions were that the greatest problems would be presented by lamb carcasses because of the high level of contamination of the fleece. Subsequent trials studied the effect of spray treatment on lamb carcasses (Bailey, 1971; 1972). Three types of spray jet were studied, all with the same pressure/flow rate characteristics, two line pressures, and two water temperatures (see Appendix A). The sprays were operated 0.3 m from the carcass surface, giving surface impact temperatures no higher than 63°C when water was supplied at 90°C. The results showed that hot water sprays were more effective than cold, though overall bacterial counts were not significantly reduced by any of the regimes studied. Hot water imparted a slight 'milkiness' initially to the surface of the carcass, which diminished on cooling and was virtually undetectable after 24 hours.

A similar manual wash was also evaluated by Kelly et al. (1981; 1982). Temperature had a far greater effect on reducing bacterial numbers than spray pressures. Kelly et al. (1982) found that although spray treatment at 80°C, as opposed to 10°C, reduced initial bacterial numbers, the storage life of hot water sprayed carcasses was no greater than that of cold water treated or cloth cleaned carcasses. The failure of the hot treatment to significantly affect the number of bacteria on the diaphragm was suggested as one reason for this. Sheridan (1982) concluded that generally manual spray treatment

systems, such as the UK and Irish systems, were impractical under commercial conditions, mainly because of speed, but also because of unacceptable carcass weight increases as a result of retention of wash water. Also the cost of using hot water sprays was considered to be excessive.

2.2.2. Spray cabinets

Extensive trials to design an automated beef carcass spray treatment unit were conducted by Anderson and co-workers at the United States Department of Agriculture Agricultural Research Service (USDA-ARS) and the Department of Food Science and Nutrition at the University of Missouri. From this work, a commercial in-line spray treatment/sanitising system, the Carcass Acquired Pathogen Elimination Reduction (CAPER) system was developed. Initial investigations into the physical factors that affect reduction of microorganisms from surfaces of red meats were carried out by Anderson et al. (1975a, b). They found that the reduction of yeasts from strips of beef improved with increasing water pressure and rate of flow; however, so did water absorption. The results supported the hypothesis that the percentage of yeasts reduced was a function of the force applied per unit of surface area. Another variable was found to be the speed of travel of meat through the spray. Based on analysis of data, equations were developed to predict the amounts of yeasts reduced on surfaces of meat and the amount of water absorbed by the meat.

Following these studies the researchers (Anderson et al., 1975a, b) concluded that a carcass decontamination treatment operation should be conducted in two different steps as each step has a different objective. The objective of the first cold washing step is to remove foreign material such as hair, dirt particles, etc.; a second sanitising procedure with hot water is then directed toward reducing the microbial population and maintaining it at a low level. Subsequently investigations concentrated on the development of an automated cleaning and sanitising unit for carcasses. Initial studies on a prototype unit looked at microbial reductions, comparing washing with combined washing and sanitising treatments; removal of foreign material; water uptake; shrinkage; carcass yield; and the effect of treatment on meat acidity (Anderson et al., 1979). Further studies investigated the problems of air flow and water loss within the cabinet system (Anderson et al., 1980; 1982a); effect of nozzle configuration and size on water absorption (Anderson et al., 1980; Anderson et al., 1982b); and cleaning-in-place (CIP) (Anderson et al., 1982c). Design specifications of a commercial CAPER system unit were described by Anderson et al. (1984), the chamber included both an initial treatment unit and a final sanitising unit using water sprays incorporating a sanitising agent (see Appendix A).

A study by Crouse et al. (1988) on the system indicated that water spray pressures of between 2412 to 4134 kNm^{-2} and chain speeds between 3.9 to 7.9 m/min had no residual effect on water uptake of the carcass after 20 hours. Also, any combination of these pressures and chain speeds reduced bacterial contamination. De Zuniga et al. (1991) in studies using a model CAPER system found no significant differences due to effect of pressure on the reduction of *Enterobacteriaceae* from the meat surface. With aerobic counts, spraying at the highest pressure, 6200 kNm^{-2}, removed less bacteria than a line pressure of 2070 kNm^{-2}, prompting investigations into bacterial penetration. Investigations into bacterial penetration into meat during treatment using Blue Lake, an insoluble dye, have been conducted by De Zuniga et al. (1991) and Anderson et al. (1991; 1992).

Graham et al. (1978) evaluated a fully enclosed spray cabinet system incorporating the specifications reported in Appendix A. This proved highly successful in laboratory trials, and in a commercial situation proved capable of handling over 300 sheep carcasses per hour and mean reductions of 2.5 log units were achieved for both total aerobic and coliform counts. Treated carcasses had a slightly cooked appearance, but reverted to normal after overnight chilling at 1 to 4°C. There were no significant differences in weight losses between treated and control carcasses.

Powell and Cain (1987) adapted this system to a beef slaughter line with the specifications reported in Appendix A. Surface impact temperatures were approximately 7°C below that of supply temperatures

at the maximum throughput studied (135 carcasses/h). At slower rates the temperature difference was smaller. Carcass size also had an effect on surface temperature.

Relatively few studies have been published on hot water spray treatment of poultry. Thomson et al. (1974) spray treated chicken carcasses with water at 20 or 30 psi at 21.1, 34.4, 60.0, 65.6 and 71.1°C. The numbers of surviving microbes decreased as the temperature of the water increased. Water at 65.6°C or higher was required to reduce the mean bacterial count by about 1 log unit. There were no significant effects of water pressure. Berrang et al. (2000) investigated spray at 73°C (on the carcass) for 20 s 30 min after defeathering, and spray at 71°C (on the carcass) for 20 s immediately after defeathering. Overall, neither of the treatments produced significant reductions in *Campylobacter*, coliform or *E. coli* counts. Li et al. (2002) did find a significant effect of using hot water in an inside-outside bird washer. Water spray treatments of 55 and 60°C for 12 s reduced inoculated numbers of *C. jejuni* by at least 0.78 log units per carcass compared to a 20°C treatment. The skin colour of the chicken carcasses was not found to be significantly effected by treatment temperatures of below 60°C.

2.2.3. Patents

Hot water-based spray processes for carcass meat treatment are described in various patents, including the following:

- C. P. Equipment in 1975 (Bliss ,1975) filed a patent for a commercial spraying apparatus designed to meet the manual spraying specification developed by Bailey (1971; 1972)
- Anderson et al. (1982) describes an oscillating system of sprayers for treatment of carcasses (U.S. Patent No. 4,337,549).
- Harben (1989) describes a hot water scalding spray for poultry (U.S. Patent No. 4,868,950).
- A patent (International Patent Number WO 02/39821 A1) by Teilmann (2002) was issued for a system to treat carcass halves with hot water in a cabin with jet devices and without spray nozzles.
- A Chad Company spray cabinet system by Anderson and Gangel (1999) describes a two-step process for treatment of carcasses before chilling which involves hot water (74°C) spraying followed by an acidic spray application.

2.3. Deluge cabinets

An alternative approach to spraying is to use a deluge, or waterfall, method of hot water distribution.

Such a system was developed in laboratory evaluations by Davey and Smith (Davey, 1988, 1989a,b, 1990a,b; Davey and Smith, 1989). This new system was compared directly with the spray cabinet used by Powell and Cain (1987) and studies showed that greater reductions could be obtained with the deluge system. The deluge system was also shown to be the more cost effective system (by a factor of 3), in terms of both achieving bacterial reductions and lower capital costs. A predictive model for processing conditions was developed based on the pilot data (Davey, 1989a,b; Davey, 1990a,b). Predictions indicated that, for a production rate of 135 sides/h, a 1 and 2 log units reduction in bacteria could be achieved at the site of minimum treatment (the neck); with meat surface temperatures of around 65°C and 80°C, respectively; at added costs of less than 0.016% and 0.03%, respectively, of the value of the meat. It was reported in 1995 (Anon, 1995) that in response to the USDA's proposal to introduce a mandatory antimicrobial treatment before chilling carcasses, the Australian Meat Research Council had funded Australian Meat Technology to develop the deluge system commercially. A cabinet was reported to have been undergoing on-line trials in a Queensland export abattoir and that evaluations during full commercial operations indicated that water could be recycled "while still retaining potability, and satisfactory organoleptic and microbiological quality" for both the

water and the meat. It was reported that a medium-sized abattoir would use about 40 litres of water per carcass. Operating at 79°C (surface temperature of about 76°C), and a contact time of about 17 s, average reductions of 2 log units reduction in *E. coli* numbers were quoted. Formal approval for recycling the water was to be sought from the US-FSIS if the evidence was satisfactory. It is unknown whether this occurred.

A system similar to the cabinet developed by Davey and Smith was used by Gill *et al.* (1995; 1997) for treating pork carcasses before evisceration but after singeing and polishing. Since dehairing and polishing causes distributes microorganisms over the whole surface of the carcass, the unit was designed to deliver sheets of water from both above and below. Initial abattoir trials (Gill *et al.*, 1995) were carried out at a slaughter plant with a throughput of about 4000 pigs a day. Carcasses were subjected to water at temperatures between 60°C and 90°C for treatment times between 20 s and 90 s. From the results of the trials, it was concluded that water at 85°C for 20 s was the most efficient treatment reducing total numbers of bacteria by about 2 log units. Subsequent trials subjecting 800 carcasses to this treatment, under otherwise normal production conditions, showed that the treatment consistently reduced numbers of spoilage organisms and *E. coli* by 2.5 log units over the whole surface of each carcass. Overall, results were very similar to those of Davey and Smith. No mention was made of high temperatures having any detrimental effects on carcass appearance in this study, but subsequent work noted that the appearance of cut muscle surfaces was permanently changed. Detailed work on pre- and post-rigor pork and beef showed that while pre-rigor beef will recover to a certain extent from heat damage, pre- and post-rigor pork will not (Gill and Badoni, 1997b). Subsequently, a fully-commercial version of the apparatus was constructed and further trials (Gill *et al.,* 1997) assessed the adequacy of the equipment under commercial conditions over three months. Carcasses were processed at rates between 600 and 800 carcasses h^{-1} with each carcass being subjected to hot water at 85°C for 15 s. An assessment of the dressing process showed that much of the subsequent *E. coli* contamination occurred during the operation for opening the throat and the floor of the mouth (Gill et al., 1997). Further work (Gill and Jones, 1998) indicated that pasteurisation (85°C for 30 s) should be delayed until after this operation. This reduced counts by around 2 log, in comparison with untreated carcasses. However, the treatment bleached the exposed muscle along the edges of the cut lines.

2.3.1. Patents

Hot water-based deluge processes for meat carcass treatment are described in various patents, including the following:

The deluge system developed by Davey (1990b) is described in the World Patent WO8901738 and U.S. Patent No. 4,965,911. The system used by Gill et al. (1995, 1997) is described in the U.S. Patent No. 5,651,730 by McGinnis et al. (1997).

2.4. Immersion

In general immersion systems have been more readily applied to poultry carcasses rather than red meat carcasses, although since pig carcasses are regularly scalded in tanks systems the technology certainly exists for treating red meat carcasses, although immersing a beef side may not be practical.

Smith and Graham (1978) found immersion in hot water to be a very effective method of reducing bacteria on the surface of meat. Immersing whole sheep carcasses in water at 80°C for 10 s reduced bacterial numbers by more than 1 log unit CFU/cm^2. Although the carcasses had a 'cooked' appearance immediately after treatment, normal surface colour returned almost completely during storage overnight in a chiller. In trials by James et al. (2000), immersion in water at 90°C for 8 s and treatment with condensing steam at 100°C produced very similar reductions. Neither system was optimised, but it was suggested that agitating the water would have probably improved the immersion treatment. The authors concluded that inherently the use of atmospheric steam was the more attractive option for commercial use, due to its simplicity. They considered that commercial application of the

immersion system presented a number of problems in terms of filtering, recirculating the water, and final disposal of the water. The buoyancy of the lamb carcasses also posed a particular practical problem.

Decontaminating poultry meat by using immersion methods has been studied by a number of researchers (Dawson et al., 1963; Pickett and Miller, 1966; Avens and Miller, 1972; Teotia and Miller, 1972; Cox et al., 1974; Thomson et al., 1979; Berrang et al., 2000; Purnell et al., 2004), with some degree of success. As would be expected, it appears from the literature that treatments are more effective at higher temperatures and with longer immersion or exposure times. Although substantial reductions may be achieved with water temperatures exceeding 60 – 65°C, such treatments can result in a partially cooked product, with browning of the flesh and tightening of the skin of the carcass.

Berrang et al. (2000) investigated the effectiveness of using hot water immediately after defeathering, i.e. essentially a second scald, for "*NY dressed*" chickens (i.e. carcasses pre-evisceration: plucked carcasses with the head, feet and entrails intact). Immersion and spray (see spray section) treatments were investigated. The immersion treatments were 1) immersion at 60°C for 28 s 30 min after defeathering, and 2) immersion at 60°C for 28 s immediately after defeathering. Overall, none of the treatments produced significant reductions in *Campylobacter*, coliform or *E. coli* counts.

Purnell et al. (2004) utilised an experimental in-line processing unit based on a scalding tank that was evaluated in a commercial poultry plant. Treatment at 75°C for 30 s significantly reduced aerobic plate counts (APC) and counts of Enterobacteriaceae and *Campylobacter*, but the skin tended to tear during trussing. However, treating carcasses at 70°C for 40 s, followed by a 12 – 15°C spray-chill treatment for 13 s, did not detrimentally affect the skin. Microbial counts remained significantly lower than the controls for eight days under typical chill-storage conditions. In further studies a more compact batch immersion system was utilised (James et al., 2000; Corry et al., 2007). In experimental studies whole chicken carcasses, inoculated with ca. 6 log unit *C. jejuni* and *Escherichia coli* K12, were treated with hot water in a pilot immersion system for 20-30 s at 75 and 80°C. A reduction of 1.3 log CFU/cm^2 in counts of *E. coli* was achieved using a 20 s, 80°C treatment. A 1.66 log CFU/cm^2 reduction in *C. jejuni* AR6, was achieved by a 30 s, 75°C treatment. Trials in a commercial poultry plant using naturally contaminated carcasses used a 20 s in hot water at 80°C. The appearance of the treated carcasses was assessed visually at intervals until the end of shelf-life, and checks made for pseudomonads, Enterobacteriaceae and *Campylobacter* on breast skin. Initial levels of *Campylobacter* spp. were low (~1 log CFU/cm^2) and variable. Numbers of *Campylobacter* were reduced, but not eliminated. Reductions of about 2 log units were obtained in aerobic counts. Visual assessment indicated that the hot water treatment caused less change in appearance than a steam treatment. It was concluded that carcasses treated with either hot water or steam could easily be used for the production of 'skin-off' portions. It was considered that changes to appearance of skin-on carcasses or portions would be acceptable to many consumers.

2.4.1. Patents

Hot water-based immersion processes for meat carcass treatment are described in various patents, including the following:

Buhot et al. (2000) patented a hot water (e.g., 80°C) process for decontamination of carcasses by immersion. This system is designed to rapidly fill and then empty a chamber once the carcass has been treated (a treatment time of 10 s is quoted). The carcass is conveyed into and out of the chamber through openings at either end of the chamber. The water is renewed for each carcass treatment. It is claimed that turbulence caused by introducing the water enhances physical treatment. The type of carcass is not detailed but the patent implies that this process could be suitable for red meat carcasses.

Schaefer et al. (2006) patented (U.S. Patent no. 7,108,882 B2) a water bath based process for continuous surface treatment of animal tissue of approximately 1 to 40 cm in length by exposure to 80-150°C for 25-150 s.

3. Studies on the efficacy of hot potable water decontamination

3.1. Immediate effect of hot potable water decontamination on the microbial load of red meat and poultry carcasses and parts

The use of hot potable water as a meat decontamination technology has been studied extensively during the last 30 years and it is now routinely used in meat processing plants in some countries. An increased number of both laboratory scale and commercial plant evaluation studies on the efficacy of hot water application in removing physical/visible contaminants such as soil, feathers and other debris and reducing the numbers of hygiene indicators (TVC, Enterobacteriaceae) and pathogenic bacteria present in meat and poultry carcasses and cuts or portions are available in the literature.

In early studies, Smith and Graham (1978) found that immersion in hot water at 80°C for 10 s resulted in l-3 log-cycle reduction in counts of *E. coli* and *Salmonella* on beef and sheep carcasses. Australian researchers (Davey, 1989a,b; 1990a,b; Davey and Smith, 1989) designed a deluging hot water treatment cabinet for decontamination of beef carcass sides by cascading a wall of hot water from low-pressure nozzles. Evaluation of the cabinet at 83.5°C for 10 s reduced counts of *E. coli* by 2.2 log-cycles, without causing permanent defects in carcass appearance (Davey and Smith, 1989). More recently, industry monitoring data from Australia indicated that beef carcasses from slaughter plants using hot water decontamination had lower average prevalence of *E. coli* (Kiermeier et al., 2006).

A pilot plant study by Barkate et al. (1993) found that spraying carcasses with hot water (95°C) for 10 s caused reductions of 1.3 log CFU/cm^2 in APC as the carcass surface temperature increased to 82°C. Gorman et al. (1995a,b) reported that spraying was more effective in reducing *E. coli* contamination on beef tissue when the pressure and temperature of the water were increased from 2.76 to 18.89 bar and from 16 and 35 to 74°C, respectively. Among other treatments, a study under commercial conditions by Reagan et al. (1996), involved hot (74.0–87.8°C at the pipe) water spraying of beef carcasses in two beef slaughtering plants for 11–18 s and with pressures of 1310–2413 kPa, resulting in reductions of APC and *E. coli* counts at the rump site by 2.0 and 1.8 log CFU/cm, respectively. Based on standard deviations, it was speculated that hot water may have achieved more consistent decontamination compared to knife-trimming of visibly contaminated tissue. Another field study by Graves Delmore et al. (1997) found that hot water rinsing (77°C, 138–152 kPa, for 2.5 or 8 s) of carcasses in one beef slaughtering plant reduced mean coliform counts by 1.3–1.8 log CFU/cm^2.

A two-step hot water treatment procedure was evaluated by Castillo et al. (1998) on beef carcass tissue contaminated with inoculated bovine faeces. First, the process involved a low-pressure treatment (25°C, 0.7 bar, 90 s) with a hand-held apparatus; then the product was exposed to warm (35°C) water for 9 s at variable pressure (17.2–31.0 bar). This step was followed by hot water treatment (95°C, 1.7 bar, 5 s). The results indicated reductions in *E. coli* O157:H7, *S.* Typhimurium, APC, and thermotolerant coliform counts by 3.7, 3.8, 2.9, and 3.3 log CFU/cm^2, respectively (Castillo et al., 1998).

Figure 1 (Appendix B) presents the effect of water temperature and exposure time on the reduction of coliform numbers after hot water spray treatment of artificially contaminated pork carcasses based on data of Castelo et al., (2001). As it is shown both water temperature and exposure time are linearly related to bacterial reduction with correlation coefficients (r^2) of 0.995 and 0.999, respectively. Similar results have been reported by Davey and Smith (1989) who found a linear relationship between water temperature and *E. coli* numbers on beef carcasses treated by spraying with hot water.

In the United States, hot water spraying is used in commercial operations to wash/treat whole beef carcasses immediately after hide removal and before evisceration in order to prevent strong attachment of contamination (Byelashov and Sofos, 2009). It was determined by Bosilevac et al. (2006) that hot water treatment (74°C, 5.5 s, 48.3 bar) of beef carcasses before evisceration in a large (380 carcasses per hour) meat packing plant reduced the prevalence of *E. coli* O157:H7 by 81% and APC and Enterobacteriaceae counts by 2.7 log-cycles (Bosilevac et al., 2006). However, according to Cabedo et

al. (1996), hot water (74°C) spraying was more effective than warm water (35°C) or chemical solutions even when applied 2 or 4 h after exposure of the tissue to contamination. It has been proposed that, in addition to killing bacteria, exposure to warm water reduces the surface tension of the water, which may enhance bacteria and fecal removal (Keener et al., 2004). Hot water (≥74°C) is considered a more effective decontamination intervention for carcasses because, in addition to a washing effect, it also causes microbial cell death and injury (Byelashov and Sofos, 2009).

Hot water has also been evaluated for decontamination of beef trimmings used for manufacturing of products such as ground beef. Gill and Badoni (1997a) concluded that immersion for 15 s min in water of 85°C reduced counts of non-thermotolerant bacteria on beef trimmings by about two log units, and that thermal pasteurization of manufacturing beef could be a commercially useful technique for reducing enteric bacteria on massively produced hamburger patties. As indicated by Ellebracht et al. (1999) hot water of 95°C for 3 s reduced *E. coli* O157:H7 and *S.* Typhimurium by 0.5 and 0.7 log CFU/g, respectively.

According to Hugas and Tsigarida (2008), the effectiveness of hot water carcass decontamination interventions depends on operational and product-related factors. Operational factors may include water temperature, pressure, flow rate and target surface distance, method of application, time or stage of application in the slaughtering process, and plant design and its operational characteristics. Intrinsic and implicit factors related to the product itself, such as animal lots, type of meat tissue, initial microbial load, the type of the microbial ecology of the product, and the time of exposure to contamination affect bacterial attachment and biofilm formation, and thus, influence the effectiveness of water decontamination treatments (Hugas and Tsigarida, 2008). As indicated by Hugas and Tsigarida (2008), the efficacy of hot water treatments on poultry decontamination may be affected by the technique used to treat carcasses. The point of application during processing (e.g., after defeathering, after evisceration or before chilling), seems to have great impact on the effectiveness of the treatment. Exposure of poultry carcasses to a second scalding after defeathering (60°C for 28 or 73°C for 20 s), did not cause additional reduction on *Campylobacter* counts compared to the first scalding (Berrang et al., 2000). Spraying poultry carcasses with hot water (55 and 60°C) in an inside–outside bird washer after evisceration and before chilling reduced *Campylobacter* counts by 0.78 log CFU/carcass compared to spraying with 20°C water (Li et al., 2002). Treatment with warm water (40°C to 70°C) may be used by poultry processors in the United States for carcass preconditioning before further treatment (Keener et al., 2004).

As indicated by Hugas and Tsigarida (2008), an alternative thermal treatment is the application of steam, which has a greater heat capacity than the same amount of water at the temperature of boiling. In addition, especially when under pressure, steam can penetrate cavities, crevices and feather follicles (James et al., 2007). Steam treatments at atmospheric pressure of poultry carcasses for up to 20 s have been shown to significantly reduce numbers of *Campylobacter jejuni* and *E. coli*, but some surface appearance damage has been observed (Whyte et al., 2003; James et al., 2007). A patented pressurized steam process for carcasses (Frigoscandia Steam Pasteurization System™) consists of an entrance section where air is blown over the sides of beef to dry surface moisture remaining from carcass treatment; this is followed by the pasteurization chamber, which is sealed and filled with steam under pressure (105°C); and an exiting section where the beef sides are sprayed with cold water (Sofos and Smith, 1998). The system is approved and used commercially in the United States.

An overview of the available data on the effect of hot water decontamination on the microbial load of beef, pork, sheep/lamb and poultry carcasses and parts is presented in Tables 1, 2, 3 and 4, of Appendix B, respectively. The results show a reduction range for APC of 0.0-2.9 log units for beef, 0.3-2.3 for pork, 0.1-3.3 for sheep/lamb, and 0.3-3.3 for poultry carcasses and parts. Similar results have been reported for other spoilage or pathogenic bacteria. For example in beef carcasses and parts the maximum reductions observed for *Salmonella*, *E. coli* O157:H7 and *Listeria monocytogenes* were 3.7, 3.8 and 2.9 log units, respectively.

3.2. Residual effect of hot potable water decontamination on microbial growth during storage of meat and poultry carcasses and parts

The available studies on the residual effect of hot potable water decontamination on microbial growth during product storage are very limited. It should be noted, however, that unlike chemical treatments, thermal treatments have no major residual antimicrobial activity during subsequent product storage. In contrast, the initial longer lag phase due to cell injury may be succeeded by rapid growth due to elevated surface tissue moisture and potential release of nutrients due to the hot water treatment (Ikeda et al., 2003; Koutsoumanis et al., 2004). Koutsoumanis et al. (2004) reported that hot water decontamination (75°C for 30 s by immersion) of beef cuts resulted in an increase of the lag phase of *L. monocytogenes* during storage at 4 and 10°C. However, growth of *L. monocytogenes* was generally faster in hot water decontaminated samples compared to untreated meat. The faster growth of the pathogen in hot water treated meat has also been observed under anaerobic conditions (Ikeda et al., 2003), and could be attributed to the potential chemical changes caused by the high temperature, which may stimulate growth of the pathogen through the increase of nutrient availability. Another possible reason could be the delay in growth of spoilage microorganisms, especially pseudomonads, caused by the heat shock which could have resulted in a decreased competition that favoured growth of the pathogen during the first stage of the storage period (Ikeda et al., 2003). The above results, however, have been observed in laboratory-scale experiments with meat cuts and need to be validated at a commercial scale on carcass decontamination. Dorsa et al. (1997) reported that spot cleaning/decontamination treatment of carcasses with hot water/steam-vacuuming and spraying with 74°C water reduced certain bacterial counts by 2.7 log CFU/cm^2; but, by 14 days of storage, counts had increased to at least 7 log CFU/cm^2. Gill and Badoni (1997a) found that pasteurization of manufacturing beef to improve the microbiological quality of ground beef provided a product of acceptable appearance and better stability during storage under a modified atmosphere and subsequent display in air.

3.3. The role of hot potable water decontamination in the compliance with the EU Process Hygiene criteria

Process Hygiene Criteria (PHC) for carcasses in the EU are laid down in Regulation (EC) No 2073/2005 and its amendments. PHC indicate the acceptance functioning of the production process and they set indicative contamination values above which corrective actions are required in order to maintain the hygiene of the process in compliance with EU food law. The use of hot water decontamination can lead to a significant improvement in the compliance with the PHC. Figures 2a and 3a show the operating characteristic curves for the EU PHC for Total Viable count (TVC) and Enterobacteriaceae in Steer/Heifer/Cow/Bull carcasses, respectively. According to these curves the effect of hot water decontamination treatment on the compliance with the PHC can be estimated based on its effectiveness in reducing the levels of TVC and Enterobacteriaceae. For example for a mean TVC concentration of 6 logs CFU/cm^2 in untreated carcasses, the applications of hot water decontamination treatment resulting in reductions of 1, 2 and 3 log units will increase the probability of acceptance from 0.009 (in untreated carcasses) to 0.522, 0.988 and 1.000, respectively (Figure 2b). Similarly, for a mean Enterobacteriaceae concentration of 4 logs CFU/cm^2 in untreated carcasses, application of hot water decontamination resulting to reductions of 1, 2 and 3 log units will increase the probability of acceptance from 0.001 (in untreated carcasses) to 0.113, 0.879 and 1.000, respectively (Figure 3b). Consequently, effective hot water decontamination can allow meat operations to meet the regulatory performance standards.

4. Studies on the efficacy of recycled hot water decontamination

Considering concerns about water resources, reconditioning and reuse of water in various types of applications, including decontamination, is of great interest. Decontamination cabinets using recycled hot water have been developed in Australia, Canada, United States and Denmark. Therefore, settling,

filtration and decontamination (e.g., chlorine, heat) systems are being developed as adjuncts to carcass decontamination technologies (Sofos and Smith, 1998).

The available data on the efficacy of recycled hot water decontamination treatments are very limited. A fully commercial hot water decontamination apparatus was installed and tested in a pork packing plant by Gill et al. (1997). The patented apparatus (Int. Patent application no. PCT CA95/00026) was designed to apply a 15 s treatment to carcasses at 0.6 m spacings, which were processed at a rate of 1200 carcasses/h. A complete description of the apparatus has been presented by Gill et al. (1997). Gill et al. (1995) evaluated the efficacy of a novel wash-pasteurization cabinet fitted with a water recirculation system for decontamination of polished, uneviscerated pig carcasses in a commercial processing plant. Through this system, sheets of hot water were delivered in an industrial scale cabinet at temperatures between 60 and 90°C for 20–90 s. Carcasses treated with 85°C for 20 s had *E. coli* counts reduced by 2 log-cycles. Treatment of 800 carcasses with water at 85°C for 20 s during normal production resulted in consistent reductions of 2.5 log-cycles. Routine (three-month) evaluation of the apparatus delivering sheets of 85°C water (15 s/carcass) onto polished, uneviscerated pig carcasses in a commercial factory processing 600–800 carcasses/h indicated reductions in the mean numbers of non-thermotolerant bacteria on carcasses (including coliforms and *E. coli*) of approximately 0.1 to 2.8 log units depending on the site of the carcass (Table 5 of Appendix B). The water, which was recirculated, carried <1 bacterium/ml and was generally free of fat, but it collected suspended and settling solids. The apparatus was presented as a commercially acceptable means of pasteurizing polished pig carcasses (Gill et al., 1997). The above apparatus was also tested for beef carcasses (Gill and Bryant, 2000). The reduction ranges for beef carcasses were 0.8-1.6, 1.0-2.3 and 1.1-2.8 logs/2500 cm^2 for APC, coliforms and *E. coli*, respectively. No significant differences were found between the efficacy of hot potable and recycled (using the above commercial apparatus) hot water decontamination. According to the findings of Gill et al. (1998), exposure of eviscerated pig carcasses or half-carcasses to 85°C for 10 s, as well as of skinned sheep carcasses to 83°C for 18 s, reduced coliform and *E. coli* counts by more than 2 log-cycles, while APC were reduced by more than 1 log unit. It was also reported that the treatment did not affect the overall carcass appearance, but it caused slight discoloration on cut muscle surfaces (Gill et al., 1998). Gill and Bryant (2000) indicated that the protein suspended in the circulating water will accumulate to levels that affect the appearance of carcass fat, unless the water is changed at appropriate intervals or actions are taken to remove the protein from the water or carcass surfaces. This system is part of the patent by McGinnis et al. (1997). Additional water re-use systems are included in the patents of Anderson and Gangel (1999) and Tielmann (2002).

5. Overall considerations of the efficacy of hot water carcass decontamination

5.1. Comparison of the efficacies of decontamination with hot potable water and recycled hot water

Due to the limited number of studies on the efficacy of decontamination with recycled hot water and the differences in the application conditions of the available studies it is difficult to compare the efficacy of the two technologies. Based on the limited available data a comparison between the efficacy of potable and recycled hot water in reducing bacteria counts on the surface of pork and beef carcasses for similar application conditions is shown in the Table 1. The above data show that under comparable conditions the efficacy of potable and recycled hot water decontamination does not differ markedly. However, further research with specifically designed studies is required for an appropriate statistical comparison of the two technologies.

Table 1: Comparison between the efficacy of potable and recycled hot water in reducing bacteria counts on the surface of pork and beef carcasses for similar application conditions

Microorganism	Temperature (°C)	Exposure time (sec)	Reduction (log CFU cm^{-2})			
			Potable water	References	Recycled water	References
			Beef Carcasses			
Aerobic bacteria	78-85	5.6-15	0.1-1.5 (*n=6)	Gill and Badoni (1997a); Graves Delmore et al. (1998); Gill et al., (1999); Delmore, Jr. et al. (2000)	0.82-1.56 (n=1)	Gill and Bryant (2000)
E. coli	78-85	8-20	0.0-3.3 (n=8)	Smith and Graham (1978); Smith and Davey (1990); Smith (1992)	1.1-2.8 (n=1)	Gill and Bryant (2000)
			Pork Carcasses			
Aerobic bacteria	85	8-12	1.38-1.70 (n=2)	Gill et al., (1998)	1.78-1.94 (n=1)	Gill et al. (1997)
Coliforms	76.6-85	8-12	>0.71-1.68 (n=3)	Gill et al., (1998); Castelo et al., (2001)	1.36-1.74 (n=1)	Gill et al. (1997)

*n: number of studies/experiments

5.2. Issues of hot water decontamination

Despite the generally accepted effectiveness of hot water decontamination technologies in reducing the numbers and prevalence of pathogenic and/or spoilage bacteria on meat and poultry carcasses, there are a number of concerns associated with their use. As mentioned previously, the period of time before decontamination has an important effect on bacterial attachment and biofilm formation, and, thus, decontamination treatments applied before evisceration will be more effective since bacterial attachment is still weak. Another important issue associated with the use of decontamination technologies is the potential development of stress-resistant pathogens (Sofos and Smith, 1998; Samelis and Sofos, 2003). Heat resistance constitutes an important physiological characteristic that may influence the behaviour of pathogenic microorganisms during meat processing and cooking. The potential concern for development of stress-resistant pathogens can be attributed to the 'stress hardening' and 'cross-protection" phenomenon which refers to the increased tolerance of a pathogen to a specific lethal stress after adaptation to the same or a different sub-lethal stress environment (Lou and Yousef, 1997; Samelis and Sofos, 2003). Several in vitro studies have demonstrated that adaptation of pathogenic bacteria, such as *L. monocytogenes*, *E. coli* O157:H7 and *Salmonella* to a mild stress environment results in increased survival under stress conditions that would be lethal for non adapted cells (Lou and Yousef, 1997; Samelis and Sofos, 2003). In addition to increased stress resistance, adaptation may lead to mutants with enhanced virulence (O'Driscoll et al., 1996) since microorganisms may sense the unfavourable conditions as a signal for the expression of virulence factors (Mekalanos, 1992). It needs to be noted, however, that, as indicated, the majority of the studies on the 'stress hardening' phenomenon have been performed in laboratory media and, thus, more research is needed on the investigation of stress adaptation in actual foods. Nevertheless, an evaluation of the contribution of decontamination interventions to food safety improvement should take into account the potential development of stress-resistant pathogens. It is also important to note that adaptation to food processing stresses is generally lost when the stressing pressure is removed and new generations of bacteria are derived. It should also be stressed that, irrespective of potential stress

adaptation inducement on survivors, decontamination treatments have been found, in countries where practiced, highly effective in reducing microbial contamination of carcasses, allowing in this manner meat operations to meet regulatory performance standards and industry specifications (Sofos, 2005). Proposed strategies to control stress resistance of bacteria involve the continued application of lethal levels of preservatives, or optimization of decontamination interventions, in type, intensity and sequence, to maximize microbial destruction and minimize resistance development (Shadbolt et al., 2001; Samelis and Sofos, 2003).

Additional concerns associated with hot water decontamination of meat (Sofos and Smith, 1998; Hugas and Tsigarida, 2008) may include potential penetration of bacterial cells into the muscle tissue when the treatment involves high pressure; it should be noted, however, that high pressures are avoided during application of hot water treatments because they cause rapid and extensive temperature reduction before the tissue is treated. De Zuniga et al. (1991) found that nozzle type had a significant effect on the penetration of a dye into the meat at a pressure of 6200 kNm^{-2}. High spray pressures tended to drive the dye into the meat surface. The relationship between line pressure and dye penetration was described by the equation reported in the Appendix A. The most susceptible surfaces to penetration were found to be those that had the covering membrane removed; cut tissue being the most susceptible. Penetration of the Blue Lake dye indicated that bacteria on the surface of meat may be able to penetrate to a depth of 0.25 mm without any treatment. It was postulated that this may be the depth of normal crevices in the meat tissue. Concerns associated with carcass treatment may also include potential spreading or redistribution of contamination on the carcass. These issues can be addressed by properly selecting, adjusting, changing and controlling the factors influencing the efficacy of spray-treatment/rinsing technologies. Treatments employing agents that inactivate bacteria (e.g., hot water and steam) can reduce contamination by killing cells with no concern of redistribution or spreading.

Another issue is whether hot water will adversely affect the quality of the product. The principle limit to the temperature of water that can be used is its effect on the meat. The treated meat must retain the appearance and characteristics of fresh raw meat. While data are available on the death kinetics of pathogenic bacteria, there is little information on the relationship between surface temperature and the appearance and colour of meat tissues. The data of Morgan et al. (1996a, b) show that it is possible to expose small pieces of chicken breast (10 mm x 10 mm, 50 mm long) to steam at 100°C for 10 sec, without cooking. These workers also provide a clear table of cooking times at higher temperatures. Data for lower temperatures are less clear. Some data for chicken breasts is provided by Göksoy et al. (2001) at temperatures between 50 to 100°C.

Numerous studies have shown that while heat treated meat may show an immediate appearance of heat damage it may recover during subsequent storage (Table 2). The appearance of carcass surface tissue immediately after exposure to treatments of 80°C or above may appear bleached, gray or 'cooked' to an approximate depth of 0.5 mm (Smith, 1992). Smith and Graham (1978) found that the cooked appearance of the surface of beef, lamb and mutton carcasses immersed in hot water (80°C for 10 s) disappeared almost completely within a few hours of cold storage. Similar observations were made by James et al. (2000) after immersing lamb carcasses in hot water at 90°C for 8 s. Smith and Davey (1990) reported that the use of a novel hot water cabinet to apply water of 83.5°C for 20 s over the entire surface of sides of beef caused a cooked/bleached appearance, but that the sides regained normal appearance during chilling. Exposure to temperatures above 85°C for more than 20 s may result in permanent damage of surface bloom (Davey and Smith, 1989). A slight visual discoloration observed by Barkate et al. (1993) on the carcass surface immediately after treatment with 95°C for 10 s was absent after 24 h of refrigerated storage and the normal carcass colour was restored. Gill and Badoni (1997a) immersed portions of post- and pre-rigor pork and beef in 75 or 85°C water for 5, 10, 15 or 20 s and concluded that pasteurizing treatments cannot be applied to meat without some degradation of the appearances of cut muscle surfaces. Gill et al. (1999) concluded that treatment of beef carcass sides with water of 85°C for 10 s should substantially reduce bacterial counts without unacceptable damage to the appearance of the product. According to Castillo et al. (2002), treatment carcass tissue with water of 80°C or higher did not result in permanent discoloration of the tissue

surface. In general, potential concerns associated with surface tissue discoloration have been discounted by most studies as being transitory under the practical conditions of treatment application. However, although this may not be true for all species, initial surface discolorations, caused by treatments based on high temperature, are usually unnoticed after a few hours of chilling. Lamb/mutton and beef has been reported to recover, however detailed work on pre- and post-rigor pork and beef by Gill and Badoni (1997a) showed that while pre-rigor beef will recover to a certain extent from heat damage, pre- and post-rigor pork will not. Poultry also appear to be relatively sensitive to temperatures over 60-65°C. With poultry the main effect of hot water treatments is to tighten the skin of the carcass making it difficult to sometimes truss the carcasses after chilling without the skin tearing (Purnell et al., 2004). Purnell et al. (2004) found that while an immersion treatment at 75°C for 30 s significantly reduced APCs and counts of Enterobacteriaceae and *Campylobacter*, the skin tended to tear during trussing. However, treating carcasses at 70°C for 40 s, followed by a 12 – 15°C spray-chill treatment for 13 s, did not detrimentally affect the skin. In subsequent trials a 80°C for 20 s immersion treatment, with no spray-chill treatment produced a generally acceptable carcass (James et al., 2000; Corry et al., 2007). Goksoy et al. (2001) reported that they found no hot water immersion treatment for poultry below 90°C that would reduce *E. coli* contamination without causing adverse changes in the appearance of the product.

Few studies have assessed the effect of hot water treatments on the eating quality of the meat. Gill et al. (2001) showed that the aspect of cooked patties manufactured with meat treated with water of 85°C for 60 s was not distinguished from the normal commercial product, and concluded that pasteurizing manufacturing beef with water of 85°C for 45 s could be a practicable treatment for enhancing the microbiological safety of frozen hamburger patties.

Table 2: Reported effects of hot water treatments on the appearance of meat tissues

Meat	Treated material	Application	Temperature °C	Exposure time (s)	Notes	Reference
Beef	Carcass	immersion	80°	10	Immediate 'cooked' appearance, recovery after chilled storage overnight	Smith and Graham (1978)
Beef	Carcass	deluge	83.5°	20	Immediate 'cooked/bleached' appearance, recovery after chilled storage overnight	Davey and Smith (1989)
Beef	Carcass	deluge	85°	20	Permanent damage to surface bloom	Davey and Smith (1989)
Beef	Carcass	spray	95°	10	"Slight discolouration" on carcass surface, recovery after 24 h chilled storage	Barkate et al. (1993)
Beef	Carcass	spray	85°	10	Acceptable	Gill et al. (1999)
Pork	Carcass	deluge	85°	30	Exposed muscle "bleached" along edges of cut lines	Gill and Jones (1998)
Lamb	Carcass	immersion	80°	10	Immediate 'cooked' appearance, recovery after chilled storage overnight	Smith and Graham (1978)
Lamb	Carcass	immersion	90°	8	Immediate 'cooked' appearance, recovery after chilled storage overnight	James et al. (2000)
Poultry	Carcass	immersion	60°	600	Limit for carcass appearance being acceptable	Morrison and Fleet (1985)
Poultry	Carcass	immersion	>60°	Not indicated	Partial cooking	Dawson (1963)
Poultry	Carcass	immersion	93°	15	Partial cooking	Pickett and Miller (1966)
Poultry	Carcass	immersion	71°	20	Acceptable	Pickett and Miller (1966)
Poultry	Carcass	immersion	71.7°	Not indicated	Partial cooking	Cox et al. (1974)
Poultry	Carcass	immersion	65°	180	Discoloured	Thomson et al. (1979)
Poultry	Carcass	spray	55°	12	Unaffected	Li et al (2002)
Poultry	Carcass	immersion	75°	30	Skin tearing during trussing	Purnell et al. (2004)
Poultry	Carcass	immersion	70°	40	Epidermal damage in 5-9% of carcasses	Purnell et al. (2004)
Poultry	Carcass	immersion	80	20	Some epidermal damage	Corry et al. (2007)

6. Assessment of potential microbiological risks associated with decontamination using recycled hot water

This chapter is not an exhaustive analysis of all hazards that actually or potentially can be associated with carcasses and/or water used for carcass decontamination treatments. Rather, it is a brief overview of bacterial and protozoan hazards that can be reasonably expected to occur on the surface of carcasses and/or in associated carcasses-treatment water under normal circumstances of abattoir operations, with particular attention paid to their main heat-related characteristics.

6.1. Main biological hazards associated with carcasses

Meats are important sources of human foodborne bacterial pathogens, mainly including *Salmonella, Campylobacter jejuni/coli*, *Yersinia enterocolitica,* verotoxigenic *Escherichia coli* and, to some extent, *Listeria monocytogenes*. All these may be harboured in the gastrointestinal tract of food-producing animals or in the environment and can contaminate animal carcasses through direct faecal contamination or cross-contamination via tools, machinery and aerosols, and subsequently some can be washed-off (dead or alive) into the carcass-decontamination water. Occurrence of foodborne pathogens such as *Salmonella, C. jejuni/coli*, *Y. enterocolitica* and verotoxigenic *E. coli* in fresh red meats including carcasses varies relatively widely, although most often it is between less than 1% and up to 10%, depending on a range of factors including the organism, geographical factors, farming and/or meat production and processing practices (Noerrung et al., 2009). However, on poultry carcasses, occurrences of some bacterial pathogens (e.g. *Campylobacter, Salmonella*) can be markedly higher compared to red meats (Noerrung and Buncic, 2008). Furthermore, direct or indirect faecal or soil contamination of carcasses can lead to presence of sporogenic and toxigenic bacterial pathogens such as *Clostridium botulinum*, *C. perfringens* and *Bacillus cereus*, as well as of protozoan parasites (e.g. *Cryptosporidium spp., Giardia duodenalis* and *Entamoeba histolitica*). In addition, carcasses potentially can become contaminated with water-borne pathogens such as *Legionella* and *Aeromonas* bacteria in case of use of contaminated water.

6.1.1. Non-spore-forming bacterial pathogens

Vegetative bacterial pathogens are sensitive to heat and should be inactivated at temperatures used to recondition hot water for re-use in carcass decontamination. They include the following:

6.1.1.1. Aeromonas

Aeromonads are associated with aquatic habitats and known to cause extra-intestinal diseases, e.g. wound infections following exposure to contaminated water (Hudson, 2004). The pathogenic species including *A. hydrophila, A. cavieae* and *A. veronii* are also associated with the normal faecal flora of farm animals. *Aeromonas* can grow between -0.1°C (some strains even at -2°C) and 45°C. *A. hydrophila* produces exotoxins (cytoxic enterotoxins) and endotoxins in culture media and foods, but their precise role in gastrointestinal disease has yet to be elucidated; both the organism and the toxins are inactivated by heat.

6.1.1.2. *Campylobacter*

Campylobacter are gram negative, motile, rod-shaped bacteria, and the most important species of *Campylobacter* are the thermophilic species: *C. jejuni, C. coli* and *C. lari*, the first two species causing almost all (*C. jejuni* ca. 90% and *C. coli* ca. 7%) human disease (Donnison and Ross, 2003; Nørrung et al., 2009). They require a microaerobic atmosphere (ca. 5% oxygen and 10% carbon dioxide) and multiply only at 30-45°C. A new finding is that *Campylobacter* and other pathogens can reside within amoebas and other parasites (Dahlgren et al., 2003) providing a possible explanation (the Trojan Horse phenomenon) of the survival and persistence of *Campylobacter* in water in particular at low temperatures and in other unfriendly environments. Nevertheless, *Campylobacter* spp. is relatively sensitive to heat and can be readily inactivated by pasteurisation.

6.1.1.3. *Legionella*

Legionella is an aerobic bacterium that is an intracellular parasite (in macrophage and neutrophil). It is associated primarily with water and has a capacity to adhere to metal, plastic and rubber in water systems. It grows between 20°C and 45°C (reportedly even at 50°C) but can be inactivated by heat treatment at 70°C in a matter of minutes (Muraca et al., 1987).

6.1.1.4. *Listeria monocytogenes*

The genus *Listeria* consists of at least six different species but only *L. monocytogenes* is regarded as pathogenic to humans. *L. monocytogenes* is a gram-positive rod and grows within the temperature range –0.4°C to 45°C. If present in meats, heat treatment to around 70°C-75°C will quickly kill the organism (Buncic and Avery, 2004).

6.1.1.5. *Salmonella*

More than two thousand and five hundred serotypes exist within the genus *Salmonella* and, although almost all serotypes are regarded as pathogenic, *S.* Typhimurium and *S.* Enteritidis cause 60-90% of all human cases of salmonellosis (Nørrung et al., 2009). The reason for this is that these serotypes are by far the most predominant in food animals. *S.* Typhimurium can be found in pigs, cattle and chickens while *S.* Enteritidis is mainly found in broilers and table-egg producing hens. *Salmonella* is a gram-negative rod and grows within the temperature range 5°C–46°C. If present in meats, heat treatment to around 70°C will kill the organism.

6.1.1.6. *Staphylococcus aureus*

S. aureus is a pathogenic microorganism that may cause infections as well as food borne intoxications. It is a gram-positive rod with a remarkable resistance in the environment, grows in the temperature interval 7-48°C, and is the most salt resistant pathogenic microorganism. The production of enterotoxin can occur between 10-48°C. *S. aureus* is easily killed by heating, but the toxin is heat-resistant (Hudson, 2004; Nørrung et al., 2009).

6.1.1.7. Verotoxigenic *Escherichia coli* (VTEC)

VTEC are strains of *Escherichia coli* capable of producing certain cytotoxins; some of these may also be enterohaemorrhagic (EHEC) due to additional pathogenic factors. Several serotypes of VTEC are known; however the majority of cases of human illness (including outbreaks) have been caused by serotype O157. In the last years the proportion of non-O157 infections has been increasing (EFSA, 2007). VTEC may grow down to about 8°C, and up to approximately 45°C (some strains even 50°C), and do not survive pasteurisation (Willshaw et al., 2000; Duffy et al., 2006).

6.1.1.8. *Yersinia*

Y. enterocolitica and *Y. pseudotuberculosis,* within the genus *Yersinia*, are food borne pathogens, and the former is by far the most frequent cause of yersiniosis worldwide. *Y. enterocolitica* occurs in several biotypes and serotypes, which differ in pathogenicity to humans, geographical distribution and animal reservoirs (EFSA, 2007). *Yersinia* is a psychrotrophic gram-negative rod, with a growth potential in the 0 to 42ºC range and is easily killed by heating (Nørrung et al., 2009).

6.1.2. Spore-forming pathogenic bacteria

Spore-forming bacterial pathogens that may be present on carcasses include *B. cereus*, *C. perfringens* and *C. botulinum* (ICMSF, 1998). While, as indicated, bacterial vegetative cells, including those of

these spore formers, are sensitive to heat and therefore killed rapidly when exposed to pasteurization and cooking temperatures (ICMSF, 1996), bacterial spores are characterized by their increased resistance to heat and other stresses (Sofos, 1989). Toxins produced by sporeformers such as *C. botulinum* are also inactivated when exposed to the temperatures range of 75-80°C.

6.1.2.1. *Bacillus cereus*

B. cereus is a spore-forming bacterium ubiquitous in the environment (soil, plants, animals guts). It grows aerobically and (slowly) anaerobically between 5°C and 50°C (some strains grow even at 55°C). *B. cereus* can produce two different toxins, emetic and diarrhoeal. Vegetative cells and diarrhoeal toxin are readily inactivated by pasteurisation, but the spores and the emetic toxin are heat resistant (Whyte and Wong, 2004).

6.1.2.2. *Clostridium botulinum*

C. botulinum is strictly anaerobic, spore-forming and neurotoxin-producing bacteria that may be present in the soil and guts of animals and humans. The species is comprised of a varied group of types, which differ in growth requirements for temperature, water activity, pH and heat treatment necessary for inactivation (Boerema and Broda, 2004). They can grow within the range of 3.3°C and 45°C (some types even 50°C). The mesophilic *C. botulinum* group can endure low water activity (i.e., 10% salt) and acidic pH, i.e. down to 4.6 (ICMSF, 1996). Both the vegetative cells and botulinum toxin are inactivated by heating, but the spores are heat-resistant.

6.1.2.3. *Clostridium difficile*

This spore-forming and toxigenic organism has drawn increased attention in recent years. It can cause enteritis, particularly where antibiotic treatment is involved. *C. difficile* occurs in domestic/food animals where it causes infections and DNA typing suggested a connection between animal and human infections (Rodriguez-Palacios et al. 2006). Vegetative forms are inactivated by pasteurization and the toxin is very unstable, but spores can survive meat cooking regimes.

6.1.2.4. *Clostridium perfringens*

C. perfringens is anaerobic (microaerophilic), spore-forming bacteria that may be present in the soil and guts of animals and humans. It grows in the temperature range of 15-50°C (although requires a high water activity to grow at the extreme values) and produces enterotoxin during sporulation. The vegetative cells are killed and the toxin is inactivated by pasteurisation, but spores can survive cooking for several hours (ICMSF, 1996).

6.1.3. Protozoa

Zoonotic protozoan parasites relevant to surface contamination of carcasses include *Cryptosporidium* spp., *Giardia duodenalis* and amoebae (Gracey et al., 1999). *Cryptosporidium* spp. and *Giardia duodenalis* can be present in faeces of farm animals (particularly in cattle) and man and cause diarrhoeal disease especially in immuno-compromised and young categories. Consequently, due to direct or indirect faecal contamination, they can be present on carcasses as well as in contaminated water. Their cysts are relatively resistant to many environmental and disinfecting factors, especially those of *Cryptosporidium*, but are inactivated by pasteurising heat treatment. *Entamoeba histolytica* is primarily parasite of man but occasionally can infect animals. It can exist as a commensal in the intestine, but also cause haemorrhagic enteritis. Similarly to *Cryptosporidium/Giardia*, amoebaean cysts can be present on carcass surface and, primarily, water due to faecal contamination, but are readily inactivated by heat (Madigan et al., 1997).

6.2. Identification of microbiological hazards of the main concern

It is assumed that all microbiological hazards present on contaminated carcasses (mentioned above) may be transferred, to some extent, from the carcass surface into the recycled hot water used for carcass decontamination. However, their further fate in the water will vary depending on a range of factors among which the inherent behaviour towards heat, i.e. the ability to multiply and/or survive at higher temperatures, is the most relevant. A summary of heat-related characteristics of most relevant microbiological hazards is presented in Table 3.

According to available information on the Danish system for recycled hot water operation, the temperature of the post-carcass-decontamination water collected in the residence tank never decreases below 75°C and is subsequently increased to at least 79.5°C before its re-use to decontaminate new carcasses. However in other systems the temperature range may differ, and should be carefully evaluated.

Based on this, none of the non-spore-forming bacteria are expected to multiply in the water collected and recycled after hot water carcass decontamination under those conditions, because none of the listed bacterial hazards has maximum growth temperature above 55°C (Table 3). Actually, such temperatures should cause vegetative cell inactivation provided that heat transfer and exposure time are adequate.

With respect to spores (not cells) of the spore-forming bacteria (*B. cereus, C. botulinum, C. perfringens, C. difficile*), it can be presumed that they, generally, can survive at the 79.5°C temperature of the recycled hot water operation described for the Danish system. Although a certain proportion of the spores may die if exposed for several hours to these temperatures (e.g., spores entering the water at start of the day), additional spores are likely to be introduced in the water throughout the day as new carcasses are processed, while dissolved/suspended proteins and fat may provide protection and enhance their survival.

With respect to microbial toxins (Table 3), the main questions are: a) whether toxins can be formed on the carcass surface and washed-off into the water used for carcass decontamination; b) whether toxins can be formed in the recycled water during the recycled hot water operation; and c) whether the toxins (if any formed) will be inactivated in the recycled water. It is reasonably assumed that the toxins are not formed on carcasses during the short period between the slaughter and the carcass decontamination treatment. Also, formation of the toxins is not expected in the recycled hot water at 79.5°C. Even in the case of hypothetical production of the bacterial toxins in the recycled water, most of them are heat sensitive and thus would be inactivated at 75-85°C – with only exceptions of heat-resistant *S. aureus* enterotoxin and *B. cereus* emetic toxin. Overall, significant presence of toxins produced by bacterial pathogens listed in the recycled hot water for carcass decontamination is not expected.

With respect to protozoa, because of their relative high sensitivity to heat (Table 3), they are not expected to survive the 79.5°C temperatures of the recycled hot water and thus presumably should not be present in that water.

Overall, based on the above considerations and assuming that the recycled water temperature never drops below 75°C (as indicated for the Danish system), microbiological hazards of main potential concern in all animal species operations are bacterial spores (e.g., *C. botulinum, C. perfringens, C. difficile* and *B. cereus*). Hence, only these microbiological hazards will be further examined in this chapter.

Table 3: Heat-related characteristics of main microbiological hazards associated with carcasses and carcass-treatment water

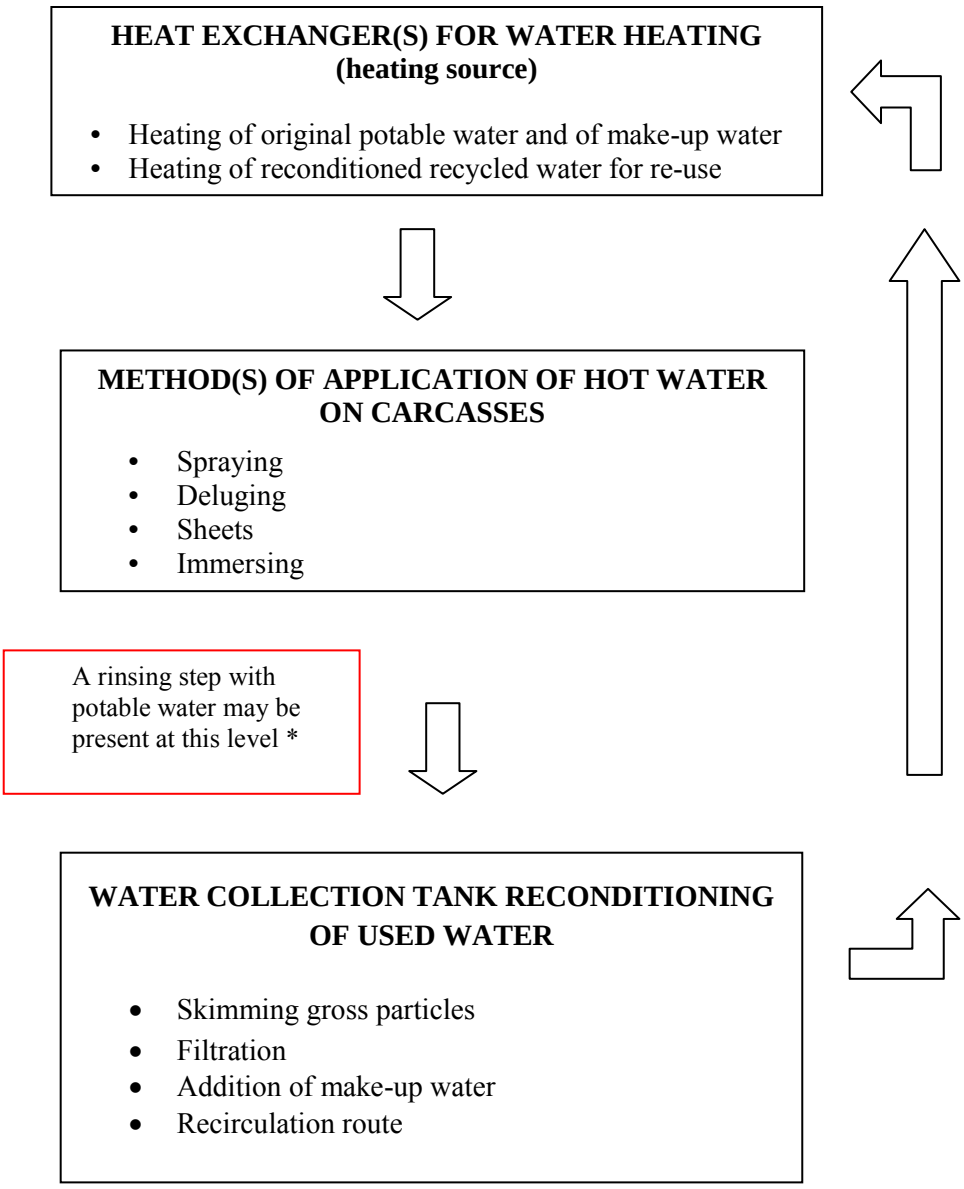
Microbiological hazards	Maximum growth temperature (°C)	Heat resistance: vegetative cells[B]	Heat resistance: spores[B]	Heat resistance: toxins	Sources (adapted from)	Survival in recycled hot water (75°C[a] to 79.5°C[b])
Non-spore forming bacteria						
Campylobacter	45	$D_{55°C}$=0.6-2.3 min	N/A	N/A	Donnison and Ross (2003); ICMSF (1996); Varnam and Evans (1991)	Negligible
Salmonella	47	$D_{70°C}$=0.07-0.39 min $D_{65°C}$ = 0.18-2.64 min $D_{60°C}$=0.2-6.5 min	N/A	N/A	Griffis and Osaili (2009); Varnam and Evans (1991)	Negligible
Verotoxigenic *Escherichia coli* (e.g. O157:H7)	45-50	$D_{70°C}$=0.030-0.08 min $D_{65°C}$=0.15-0.75 min $D_{60°C}$=0.1-2.64 min	N/A	Medium thermo-resistance	Griffis and Osaili (2009); Dykes (2004); Guerrero - Legarreta (2004); ICMSF (1996)	Cells: negligible Toxin: possible, but its formation is unlikely
Listeria monocytogenes	45	$D_{70°C}$=0.11-0.47 min $D_{65°C}$=0.3-2.21 min $D_{60°C}$=5-8.32 min	N/A	N/A	Griffis and Osaili (2009); ICMSF (1996); Varnam and Evans (1991)	Negligible
Staphylococcus aureus	48	$D_{60°C}$=0.43-7.9; but 25 min in 8.4% salt	N/A	High thermo-resistance	Hudson (2004); Guerrero - Legarreta (2004); Varnam and Evans (1991)	Cells: negligible Toxin: possible, but its formation is unlikely
Yersinia enterocolitica	42	$D_{71.8°C}$=0.33 min $D_{62.8°C}$=0.01-1 min	N/A	N/A	Mills (2004); Varnam and Evans (1991)	Negligible
Aeromonas hydrophila	41-45	$D_{55°C}$=0.2 min $D_{48°C}$=3.5-6.6 min	N/A	Low thermo-resistance	Hudson (2004); ICMSF (1996)	Cells: negligible Toxin: negligible
Legionella	45	$D_{60°C}$=3.2 min $D_{70°C}$=0.7-2.6 min	N/A	N/A	Stout et al. (1986); Schulze-Roebbecke et al. (1987)	Negligible

Microbiological hazards	Maximum growth temperature (°C)	Heat resistance: vegetative cells[B]	Heat resistance: spores[B]	Heat resistance: toxins	Sources (adapted from)	Survival in recycled hot water (75°C[a] to 79.5°C[b])
Spore-forming bacteria						
Bacillus cereus	43-55	Inactivated by pasteurisation	$D_{95°C}$=1.5-36.2 min $D_{85°C}$=33.8-106 min $D_{85°C}$=34-202 min	High thermo-resistance of emetic toxin (surviving 126°C/90 min); Low thermo-resistance of diarrhoeal (inactivated at 56°C/5 min)	Whyte and Wong (2004); ICMSF (1996); Varnam and Evans (1991)	Cells: negligible Spores: possible Emetic toxin: possible, but its formation unlikely Diarrhoeal toxin: negligible
Clostridium perfringens	50	Inactivated at >55°C	$D_{95°C}$=>200 min $D_{80°C}$=50-120 min (but outbreak strains survive >60-fold better than non-outbreak)	Low thermo-resistance of intracellular enterotoxin (inactivated at 60°C/5 min)	Broda and Boerema (2004); ICMSF (1996); Varnam and Evans (1991)	Cells: negligible Spores: possible Toxin: negligible
Clostridium botulinum groups III and IV	ND	Inactivated by pasteurisation	$D_{104°C}$=0.1-1.2 min	Low thermo-resistance (inactivated at 80°C/10 min)	Boerema and Broda (2004)	Cells: negligible Spores: possible Toxin: negligible
Clostridium difficile	at least 42	Inactivated by pasteurisation	Survive 71°C for 2 h; when reheated D85°C= 10 min	Very unstable, degrades at room temperature	Karlsson et al. (2003); Rodriguez-Palacios et al. (2010); CDC (2010)	Cells: negligible Spores: possible Toxin: negligible
Protozoan parasites						
Cryptosporidium spp., Giardia duodenalis, Entamoeba histolitica	N/A	Inactivated at >58°C	Oocysts *C. parvum* inactivated at 45°C/5 min	N/A	Madigan et al. (1997); Varnam and Evans (1991)	Negligible

N/A=Not applicable; ND=No data; B=z-values range between 4.4° and 10°C (Stumbo 1973; Adams and Moss 1995; Reichert, 1979); a=water in the resident tank; b=reheated water

6.3. Factors affecting the levels of spores in recycled hot water

There is a lack of data on the effects of various steps of the recycled hot water operation on the incidence and levels of relevant hazards (i.e., bacterial spores) in the water to be re-used for carcass decontamination. Nevertheless, the potential relevant hazards will be generally outlined (below) based on general process hygiene- and food hygiene-related principles as applied to a generic model of recycled hot water system. Components of a generic hot water recycling system



* The rinsing step after treatment with recycled hot water increases the safety level of the process

6.3.1. The effects of incidence/initial level of spores on carcasses

The prevalence of bacterial spores on carcass surfaces is a key factor determining their occurrence/levels in the recycled water. Most clostridia found in raw meats are harmless putrefactive mesophiles, while *C. botulinum* is rarely found in raw poultry, beef and pork (Johnson, 2007). Estimates of contamination levels range from <0.1 to 7 spores per kg of meat (Hauschild, 1989; ICMSF, 1998). Thirteen surveys of meat have found *C. botulinum* present at a MPN estimate of 7 spores/kg (Peck, 2009). The incidence appears to be lower in beef and lamb than in pork. *Clostridium perfringens* is commonly (30-80% of samples positive) isolated from the surface of beef, sheep and pig carcasses at slaughter, but at low numbers (<200/100cm^2) and mainly as vegetative cells (Labbe, 1989; ICMSF, 1998). According to Juneja et al. (2009) it was detected at rates of 29, 66, and 35% in beef, pork, and lamb carcasses, respectively. *B. cereus* may also be frequently isolated from meat and meat products (Griffiths, 2009). Raw meats were found positive at a rate of 6% with levels in the range of 1-2 log units per gram, while raw hamburger samples were 34% positive with 1-3 log units of contamination (Kramer and Gilbert, 1989). In general, *B. cereus* contamination levels in meat are less than 5 log units per gram.

Overall, the occurrence/level of bacterial pathogen spores on carcasses is expected to be low and highly variable, and to depend on pre-slaughter factors and the slaughter line process hygiene.

6.3.2. The effects of physical aspects of hot water application on carcasses

Unfortunately, the meat-water transfer ratios for spores are not only unknown, but they are also likely to vary with water treatment methods and carcass surface characteristics. Water pressure and total water amount per carcass are likely to influence the meat-water transfer ratios of spores, and hence to affect the occurrence/levels of spores in the recycled water. In addition, the ratio between hot water volume and carcass surface will determine the extent of "diluting" effects on the hazards in the collected water. Overall, it could be expected that the higher the pressure and the volume of water used, the higher proportion of the hazards is rinsed-off the carcass; in which case the spores removed would be diluted in more water used for recycling.

6.3.3. The effects of skimming

In the used water collection residence tank, the floating substances in the collected post-carcass-treatment hot water (e.g., fat) are skimmed off, and, thus, understandably, some spores entrapped in the skimmed materials would be removed from the water. However, the total proportion of skimmed off spores is likely to be relatively small - as the volume of the water is much larger than the volume of the removed solid material. Nevertheless the contribution of skimming in removal of potentially present bacterial spores is unknown

6.3.4. The effects of water filtration

Collected post-carcass-decontamination water is filtered to remove particles >150 µm. In principle, the filtration is not expected to remove spores because of their much smaller sizes, unless they are entrapped in the filtered residues. Nevertheless, it is possible that a proportion of the spores can be attached to the tissue particles washed-off from the carcasses, and, this, would be filtered-off from the recycled water together with them. The proportion of filtered-off spores (if any) is unknown and likely to vary with the total amount of particles in the water and the efficacy of the filtering process.

6.3.5. The effects of addition of clean water

In the Danish system, when the collected post-carcass-decontamination hot water becomes too turbid (>72 NTU), approximately 20% of clean water is added to the recycled water. Presumably, the same volume of recycled water is discarded and/or lost in the process. If such is the case, it can be assumed

that the concentration of spores in the water would be reduced by approximately 20% each time that clean water is added.

6.3.6. The effects of reheating recycled-water

The skimmed and filtered water is re-heated from presumably its lowest temperature of 75°C in the residence tank to at least 79.5°C (with the Danish system) or even up to 85°C in some other systems. The 5-10°C increase in the water temperature during the reheating step would reduce D-values, as typical z-values (10-fold reducing D-values) for the large majority of bacterial hazards range between 5 and 10°C. Because D-values of bacterial spores even at 80-85°C are in the order of hours (Table 3), the effect of reheating the water from 75°C to 79.5°C on the overall spore counts may not be necessarily very high

Bacterial spores are metabolically dormant and have remarkable heat resistance as those of some species can withstand temperatures of 100°C (Setlow and Johnson, 2007). Exposure to elevated temperatures (50-170°C) may stress and injure bacterial spores (Sofos, 1989). Heat injured spores are unable to develop visible signs of growth, under conditions that are optimal for unheated spores, exhibit sensitivities to various agents, and need special conditions for detection. Thermal D-values of spores, as reported by Setlow and Johnson (2007) are: for *C. botulinum* Group I, types A and B 50, 7-30, 1-3, and 0.1-0.2 minutes at 90, 100, 110, and 120°C, respectively; for *C. botulinum* Group II, type B: 1-30, 0.1-3, and 0.03-2 minutes at 85, 90, and 95°C, respectively; for *C. botulinum* type E: 0.3-3 and 0.01 minutes at 80 and 100°C, respectively; for *C. perfringens*: 3-145, 0.3-18, and 0.03-2.4 minutes at 90, 100, and 110°C, respectively; and, for *B. cereus*: 3-200 and 0.03-2.4 minutes at 90 and 110°C, respectively (Setlow and Johnson, 2007). Thus, temperatures in the range used to recycle hot water for treatment of carcasses will not result in killing of spores.

In order for bacterial spores to become vegetative cells, which are metabolically active, able to divide and heat sensitive, they need to germinate, which follows spore activation. In general, spores germinate more rapidly and completely if activated prior to exposure to a germinant (Setlow and Johnson, 2007; Sofos, 1989). Spore activation is generally accomplished by exposure to sublethal (below spore inactivation) heat treatments, or other treatments (e.g., low pH, alcohol). *B. cereus*, *C. perfringens* and *C. botulinum* have been reported as germinating at temperatures up to 50--80 °C (Kramer and Gilbert, 1989; Labbe, 1989; Hauschild, 1989). As indicated, activation accelerates the germination process, which makes the organism sensitive to heat. Initiation of spore germination may be triggered by exposure to various agents including amino acids, sugars and salts, some of which may be released from carcasses in the recycled hot water (Sofos, 1989), but their potential involvement is unknown.

In general, during the germination stage of the life cycle of spore-forming bacteria, the dormant state of the spore is irreversibly terminated through a complex series of events and leads to a stress-sensitive, metabolically active form, which is ready for the next stage, the outgrowth. Outgrowth is a transitional stage, during which the germinated spore is transformed into a vegetative cell. The growth stage involves metabolism and vegetative cell division. The cells produced are sensitive to heat (Sofos, 1989). Reheating water for reuse in recycled hot water decontamination systems could be considered as potentially allowing or contributing to spore activation, germination, outgrowth and inactivation resulting cells due to a form of tyndallisation[8] processing. However, no data are available to support this hypothesis.

Tyndallisation or fractional sterilization is a method developed to destroy bacteria and spores by exposure to two or three sequential heating treatments lower than those needed for direct inactivation of spores by sterilization at temperatures above 100°C. The process is named after 19th century British physicist John Tyndall. It involves heating at temperatures in the range 80-100°C for approximately

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15-60 minutes on each of 2-3 successive days; the material is kept at 30-37°C overnight between successive heating treatments. Tyndallisation is based on spore activation during heating, germination between the heating periods, and inactivation of vegetative cells during the second and third heating. Simply, the basic principle is that heat resistant spores will germinate after each heating and become susceptible to killing during the second and third heating (Gould, 2006; Alder and Simpson, 1992). Double heating in the range 89-99°C inactivated spores of *B. subtilis* more effectively as compared to single heating; the holding period used in this study was 20 minutes at 37°C (Cho et al., 1999). This, hypothesis, however, remains to be verified under temperature and time conditions involved in reheating and reusing recycled hot water for carcass decontamination. No data are available to confirm such events in recycled hot water decontamination systems.

The issue of the fate of bacterial spores during decontamination with recycled hot water is further complicated due to the unknown effects of skimming, filtration, and make-up water addition mentioned above. Furthermore, as long as the recycled hot water system is effectively cleaned and disinfected at the end of use, it is not expected that the spores will be carried over from one day to the next. Overall, if the recycled water becomes contaminated with bacterial spores either from the carcasses or the water-handling surfaces, or both, it is presumed that spores may to some degree accumulate in the water as the number of treated carcasses increases and some of those spores should be present in the recycled water used for treatment of subsequent carcasses. Whether bacterial spores should be a safety concern when exposing carcasses to recycled hot water, greater than that of when using hot potable water, will depend on the frequency of contamination, levels of spore occurrence on carcasses, degree of spore concentration in recycled water, and the fate of spores when the water is treated by skimming, filtration, make-up water addition, and exposure to 79.5°C.

However, it is also important to note that fresh meat is stored at ≤ 4°C, temperatures that are too low for growth of sporeforming bacteria. The minimum temperature allowing growth of *C. perfringens* and *C. botulinum* is 10—50°C (15°C for most strains, and 3.5°C for non-proteolytic *C. botulinum* type E strains found mostly in seafood), while the minimum temperature for *B. cereus* growth is 5 °C. Viable vegetative cells should decrease in numbers during cold storage, while cooking destroys survivors. In addition to low storage temperature, competitive psychrotrophic organisms may interfere with multiplication of spore forming pathogens in raw meat (ICMSF, 1998). It should also be noted that no cases of botulism have been documented from consumption of fresh meat, while *B. cereus* illness is mostly associated with foods of plant origin.

In general, hot water decontamination temperatures may stimulate bacterial spore activation and cause inactivation of generated vegetative cells provided that heat transfer, temperature achieved and length of exposure time are adequate (Sofos, 1989; Novak and Yuan, 2004). Considering the low prevalence and level of contamination of carcasses with spores of bacterial pathogens, that the water temperatures involved in hot water recycling could activate spores for germination and subsequently make them sensitive to thermal inactivation during water reheating, and the absence or limited growth of any surviving spores at the hot water decontamination temperatures used, the potential for increased safety risks should be negligible. Experimental data are needed, however, to document this conclusion.

6.3.7. The net result of effects of all the considered factors

Because all steps and factors associated with recycled hot water operation (indicated above) carry significant uncertainties and are variable, changeable during the day, and interfering with each other, it is expected that their net effect in terms of final concentration of spores in the recycled hot water is also variable and changeable during the day. For example, in the morning i.e. after treatment of only few carcasses, the concentration of the hazard is expectedly lower, and increases with the time, i.e. with total number of carcasses from which the heat-resistant hazards are rinsed into the water (the hazard "accumulation"). Overall, the concentration of the hazard will increase during the day if the continuous incoming hazard from carcasses exceeds the reducing effects (inactivation, filtering-

skimming removal, clean water dilution); and vice versa. Therefore, the net result cannot be quantified without knowing the quantitative contribution of each of the above-mentioned factors.

6.4. Potential for cross-contamination of carcasses via the recycled hot water treatment

Generally, the potential for cross-contamination of carcasses through the use of recycled hot water with a given microbiological hazard will depend on: a) the presence/level of the hazard in the recycled water applied to the carcass; b) the proportion of the recycled water-borne hazard that will remain on the treated carcass surface; and c) the proportion of the recycled water-borne hazard on the carcass that will be subsequently rinsed-off during post-treatment carcass rinsing with clean water (occurring with the Danish system).

6.4.1. Bacterial spores in recycled hot water used for carcass decontamination

Published data on the presence/levels of microbial hazards in recycled hot water used for carcass treatment are scarce. Gill and Bryant (2000) reported that no aerobes, coliforms, *E. coli* and clostridia were isolated from four samples of recycled hot water used for beef carcasses treatment, and the fifth sample yielded only two aerobes. Furthermore, in an Australian study (Bill and Shay, 1995) based on 10 samples of recycled hot water used for bovine carcasses treatment, "germ count" varied between 67 and 607 per 100 ml, coliform and thermotolerant coliform bacteria were not found, and from one sample a single sulphite-reducing anaerobic bacterium was isolated. On the other hand, whether and to what extent the activation/germination of spores and killing of generated sensitive cells occurs in the recycled hot water need to be considered, when data become available.

Presently, no published information on the microbiological status of recycled hot water used for the treatment of carcasses of species other than cattle and pigs is available. As indicated previously, it could be assumed that the presence/levels of the main biological hazards of concern – bacterial spores – in the recycled hot water will depend largely on their presence/levels on treated carcasses; which is variable both between animal species in a given geographical region, between same-species carcasses at different abattoirs in a given region, and between regions. Therefore, for more definite and reliable conclusion on presence/levels of the hazards (or lack of them) in the recycled hot water further related data is needed.

6.4.2. Attachment of bacterial spores from the recycled water onto the carcass surface during the treatment

Attachment of bacteria to solid surfaces including meat has been studied extensively, but the fundamental mechanisms of the attachment remain unclear (Warriner et al., 2001). With respect to bacterial attachment to carcass surface, it seems that a larger number of firmly attached bacteria is found on fatty tissue compared to lean muscle (Rivas et al., 2006), possibly because of the hydrophobic nature of the former. Furthermore, it is believed that bacteria attach better to collagen-containing connective tissues than to myofibrils (Cabedo et al., 1997; Warriner et al., 2001). During the first minutes upon arrival onto a surface, a large proportion of bacteria become attached although the whole attachment process can last over 30 min (Hood and Zottola, 1995). Nevertheless, it is considered that most bacteria are attached to meat by relatively weak forces (Warriner et al., 2001; Rivas et al., 2006). Understandably, it could be assumed that attachment of vegetative bacterial cells to meat differ from that of spores, due to their different properties and physiological status. However, no published information either on the dynamics of the attachment of clostridial spores to meat or on hot water-to-meat transfer ratio are available. Furthermore, transfer of the hazards from the water to the meat will depend on the physical parameters of the hot water treatment of carcasses (e.g. volume, pressure) and a ratio between hazards' attachment to meat and their rinsing off from it - occurring concurrently during the treatment. Overall, in cases where bacterial spores are indeed present in the recycled hot water used for carcass decontamination, it could be expected that some of them will remain on the treated carcass surface, but the extent of this cannot be quantified at present.

6.4.3. Rinsing off of bacterial spores during post-decontamination treatment of carcasses with clean water

If carcasses are rinsed with clean water following the recycled hot water carcass decontamination treatment (occurring with the Danish system), it is expected that at least a proportion of bacterial spores that had contaminated the carcass surface via the recycled hot water treatment would be removed by the clean water rinsing. It is likely that the removal would be affected by physical parameters of the rinsing treatment, as well as by the carcass surface characteristics. However, because of insufficient information on the effects of such variables, the spore-removal effect of the clean water rinsing cannot be quantified presently. Furthermore, it is unclear whether the water used for carcass rinsing is discharged or collected into the recycled water system.

6.5. Comparison of microbiological risks for carcasses treated with different decontamination systems

Three basic scenarios with respect to carcass decontamination are of interest: a) no decontamination treatments; b) decontamination with hot potable water; and c) decontamination with recycled hot water.

It has been well recognised that a certain proportion of carcasses become contaminated with microbial hazards even under best abattoir process hygiene conditions. Further improvements of the microbial status of carcasses are therefore achievable only through additional interventions such as decontamination treatments including hot water. Carcasses of all scenarios are first spray-washed with cold water

It can be presumed that the carcass status with respect to main biological hazards will improve after recycled hot water treatment, as compared with no treatment; the beneficial effects are indicated in previous chapters. However, if the recycled water contained a thermo-resistant hazard - bacterial spores – a certain degree of water mediated cross-contamination potentially of all treated carcasses with these hazards would be expected. Therefore, in the case of recycled water containing the hazards, the net meat safety effect of the recycled hot water carcass decontamination would be a function of the beneficial effects of reducing other thermo-sensitive microbiological hazards *versus* the undesirable effects of potential cross-contaminating with bacterial spores. In contrast, in cases where the recycled water does not contain these hazards, the net meat safety effect of the recycled water carcass decontamination would be only beneficial.

It can be presumed that the answer whether and how carcass status with respect to microbiological hazards will differ between hot potable water treatment and recycled hot water treatment will depend on: a) whether the decontaminating effectiveness of the two treatments differ; and b) whether hot potable water and recycled hot water differ in respect to clostridial spores. Considerations in previous chapters indicated that overall decontaminating effectiveness of the two treatments is comparable. If, at the same time, recycled hot water did not contain more bacterial spores than the hot potable water, the recycled hot water treatment would be considered equally safe (from a carcass cross-contamination perspective) to hot potable water treatment in respect to microbiological hazards. If it contained more of these hazards, the recycled hot water treatment would be considered as potentially enabling cross-contamination so comparably less safe than the hot potable water treatments. However, to definitely conclude whether recycled hot water, obtained under varying practical conditions and circumstances, contains more frequently or higher levels of bacterial spores than hot potable water (as well as if the differences are meat-safety significant), further and more detailed data are needed.

Overall, from the microbial cross-contamination of carcasses perspective, the use of recycled hot water would not increase the risk compared with use of hot potable water - as long as the recycled water has the same status with respect to bacterial spores. In other words, the use of recycled water could be allowed for carcass decontamination purpose if/where it satisfies the spore-related criteria for potable water (currently <1 spore in 100ml. The compliance of the recycled hot water with the spore-

related criteria can be ensured and verified through HACCP-based measures and external auditing (including official controls). Whether the use of recycled hot water is allowable for carcass decontamination purposes from abiotic cross-contamination of carcasses perspective will be considered in following chapters.

It is important to note that the potential appropriateness/acceptability (or the opposite) of use of recycled hot water, for purposes other than carcass decontamination, is not considered in this Opinion.

7. Assessment of the abiotic risks for carcasses subjected to decontamination treatments with recycled hot water

7.1. Main abiotic hazards associated with carcasses

Abiotic risks correspond to all risks not related to biota (living organisms). In practice this refers to the risks related to the presence of harmful chemical substances and to the possible presence of physical factors, also called "extraneous matter", such as hair, bones, or pieces of metal from the machinery present in the water.

The abiotic risk contamination for carcasses subjected to the recycled hot water treatment in slaughterhouses is mainly related to contaminants (see Glossary) such as heavy metals and other water soluble compounds such as nitrated, nitrites fluorides, as well as to organic pollutants (halogenated heterocyclic compounds) and residues of veterinary medicinal products. In addition, in rare cases radionuclides (radioactive isotopes) would have to be considered.

In summary, abiotic materials in recycled hot water can be allocated to two categories: particulate and soluble substances.

7.1.1. Particulate substances

Carcass dip water may contain small portions of animal tissues as well as potentially other extraneous matter. All technical processes include a filtration step removing particle > 150 µm, and a skimming step to remove semi-soluble fats. Hence it can be assumed that recycled water is free of any significant load of particles > 150 µm.

7.1.2. Soluble substances

7.1.2.1. Contaminants

In line with Regulation (EC) No 853/2004,[9] food business operators shall not use any substance other than potable water[10] - or, when the Regulation (EC) No 852/2004[11] permits its use, clean water[10] - to remove surface decontamination from products of animal origin (see also the Glossary). The criteria related to the quality of water intended for human consumption are laid down in Council Directive 98/83/EC.[12] Chemical parameters are detailed in Annex I, part B of the Directive.

Hot water application as a sanitizing process on large carcasses, such as bovine, porcine and small ruminant carcasses addresses particularly the muscle tissue and superficial fat and connective tissues. On theoretic grounds animal tissues may contain numerous contaminants that have been acquired in

9 OJ L139/55, 30.04.2004, p. 55–205.
10 See Glossary
11 OJ L139/1, 30.04. 2004
12 OJ L330, 05.12.1998, p. 32-54.

the live phase of the animal. Considering the short exposure time in technical decontamination processes it seems unlikely that residual amounts of these contaminants are washed out of the tissues in significant amounts. However, chemical contaminants are generally heat-stable and not be degraded by the temperature gradient in recycled hot water.

According to the above mentioned Council Directive 98/83/EC[12] the use of potable or clean water is mandatory for the use in surface decontamination. Regular chemical analysis should therefore be implemented to ensure that recycled hot water complies with these criteria. The Panel on Contaminants in the Food Chain (CONTAM Panel) noted that contaminants currently not covered by the Council Directive 98/83/EC, may also be present in recycled hot water.

7.1.2.2. Veterinary medicinal products

If hot water application as a sanitizing process is applied on poultry carcasses the potential risk of residues from veterinary medicinal products in these carcasses has to be taken into consideration. In contrast to carcasses of large animals, the kidneys or fragments of renal tissue remain in the poultry carcasses as intended for human consumption. The kidneys are a major excretion organ of an animal and may contain residual amounts of veterinary medicinal products and/or their metabolites at low levels, even when complying with prescribed maximum residue levels (MRL)[13] (see Commission Regulation (EU) No 37/2010)[14] The use of hot water will remove renal blood and cytoplasmic fluid that may contain these soluble drug residues. Of specific concern are residues of antibiotics (such as amino-penicillins, fluoroquinolones and sulfonamides and their biologically active metabolites), which may be used on large scale (treatment of the entire flock) and that are often excreted via the kidneys in a highly water soluble form.

Hence there is a risk that recycling of hot water may result in an undesirable accumulation of these heat-stable antimicrobial residues and their metabolites in this water, as poultry is slaughtered in extreme high numbers per day in commercial slaughter houses (for example 70,000 broilers per day). The CONTAM Panel recommended that regular controls for the absence of residues, particularly the absence of residues with antimicrobial activity, should be implemented

13 See Glossary
14 OJ N° L 15, 20.1.2010, p. 1-72.

8. Criteria for HACCP needed to obtain the expected efficacy and to control the possible microbiological risks

It is established and accepted worldwide that the best, currently available, system to manage processes in order to enhance food safety, based on a hazard prevention approach, is to operate under the hazard analysis critical control point (HACCP) system. According to the terms of reference of this opinion one of the tasks is to identify and define criteria for the HACCP in order to obtain the expected efficacy and to control the possible risk. Parameters and associated criteria that need to be controlled during decontamination with recycled hot water include those associated with water quality and those that need to be controlled because they impact decontamination efficacy.

Potential water quality associated criteria include:

1. Frequency and amount of make-up addition or replacement with new hot potable water, and/or suggested parameters to verify this, e.g., used water turbidity, fat modifications, denaturation of proteins, etc).
2. Abiotic particulate and soluble substances.

Potential criteria associated with decontamination efficacy include:

1. Range of water application temperature,
2. Range of water application pressure,
3. Target carcass surface temperature,
4. Length of application time (e.g., number of carcasses/hour),
5. Water flow rate,
6. Amount of applied potable or recycled hot water,
7. Frequency of recycled water replacement, and
8. Amount of make-up water used

The parameters identified as most critical to be controlled under HACCP are shown in the Table 4. The additional parameters listed above shall be considered as critical control points.

Table 4: HACCP-based control of most important parameters

Parameters to be controlled	Criteria	Critical limits	Validation	Monitoring	Corrective action	Verification
Microbiological quality of recycled water	Compliance with microbiological criteria of potable water	Temperature/time combination	Microbiological testing	Continuous recording of time/temperature	Start with new potable water; reprocess carcasses already treated	Periodical microbiological testing of water
	Heat resistant spores	No accumulation	Microbiological testing	Depending on the rate of the change of spore level	Start with new potable water; reprocess carcasses already treated	Periodical microbiological testing of water
Physical/chemical quality of recycled water	• Compliance with chemical criteria of potable water. • In case of poultry, absence of residues of relevant veterinary drugs	The same as those for potable water.	Chemical testing	Monitor scheme for potable water	Start with new potable water; reprocess carcasses already treated	Periodical chemical testing of water
Range of temperature and other parameters of recycled hot water applied on carcasses	The same as those for hot potable water	The same as those for hot potable water	The same as those for hot potable water	The same as those for hot potable water	The same as those for hot potable water	The same as those for hot potable water

CONCLUSIONS

1. Assessment of the efficacy of decontamination using recycled hot water in terms of removal/reduction of surface contamination.

- The published available data on the efficacy of recycled hot water decontamination are very limited and relate only to:

  - spray and deluge application techniques;

  - treatment of bovine and porcine carcasses.

- Nevertheless, the limited available data have shown no significant differences in decontamination efficacy, in terms of microbial reductions achievable on carcasses, between hot potable and hot recycled water, by using the recycling operations considered in this document.

2. Evaluation of the microbiological and abiotic risks for the carcases arising from the carcases and/or the water system when using recycled hot water and suggest control options.

Microbiological risk

- Application of proper heating regime of recycled water is the main option to control vegetative bacterial cells and protozoan parasites; microbial toxins are not significantly produced and/or are inactivated in the recycled hot water.

- Assuming proper heating regime, the main potential microbiological risks in the recycled water derive from heat-resistant bacterial spores such as *C. botulinum*, *C. perfringens*, *C. difficile* and *B. cereus*. However, there is a lack of data on the extent of carcass contamination with spores, their germination and inactivation during the recycling process, and the potential for accumulation during the operations.

- The control option for spores is to define a proper criteria for the HACCP in order to ensure that the microbiological risk in recycled water is not higher than in hot potable water.

Abiotic risk

- If compliance of recycled hot water with the existing chemical criteria for potable water is ensured, it is unlikely that there would be an increased abiotic risk using recycled hot water for decontamination of carcasses as compared to hot potable water decontamination treatment. However, the existing criteria for potable water neither include all chemical contaminants nor veterinary medicinal products, which might contaminate recycled hot water.

3. Identify and define criteria for the HACCP in order to obtain the expected efficacy and to control the possible microbiological and abiotic risks:

- Minimal heating temperature/time regime for recycled water before its application on carcasses must ensure compliance with existing microbiological criteria for potable water. In addition the level of heat resistant bacterial spores has to be verified by periodic monitoring. Different time/temperature heating regimes and frequency of renewal of water can be used, if microbiologically validated, continuously monitored by instrumental measurements, verified periodically by microbiological testing of water and documented.

- For recycled hot water applied on carcasses, different temperatures and application techniques-related parameters can be used, but their critical values have to be specified, validated, monitored, verified and documented in the same way as with hot potable water decontamination.

- Compliance with the chemical criteria for potable water need to be verified for recycled hot water by periodic chemical analysis of the water, and documented. In addition, the absence of residues of veterinary medicinal products in the recycled hot water used for decontamination of poultry carcasses has to be verified by periodical testing, and documented.

RECOMMENDATIONS

- Further research should be encouraged on:

    - Presence and potential accumulation of bacterial spores in the hot recycled water for decontamination of carcasses of all animal species;

    - Potential presence and accumulation of residues of veterinary drugs and other chemical contaminants not addressed in Council Directive 98/83/EC in the hot recycled water for decontamination of poultry carcasses.

REFERENCES

Acuff GR, 2005. Chemical decontamination strategies for meat. In: Improving the Safety of Fresh Meat. Ed Sofos JN, Woodhead Publishing Limited, CRC Press, New York, 351-363.

Adams MR and Moss MO, 1995. Food microbiology. Royal Society of Chemistry, Cambridge, 201pp.

Alder VG and Simpson RA, 1992. Heat sterilization. Sterilization and Disinfection by Heat Methods. In Principles and Practice of Disinfection, Preservation and Sterilization. Eds Russell AD, Hugo WB and Ayliffe GAJ. Blackwell Scientific Publications, London, 483-498.

Anderson ME, Marshall RT, Naumann HD and Stringer WC, 1975a. Physical factors that affect removal of yeasts from meat surfaces with water sprays. Journal of Food Science 40, 1232-1235.

Anderson ME, Marshall RT, Naumann HD and Stringer WC, 1975b. Design criteria for spray cleaning of beef - a model system. Presented at the 1975 Annual Meeting of the American Society Of Agricultural Engineers. Paper No. 75-6010.

Anderson ME, Marshall RT, Stringer WC, and Naumann HD, 1979. Evaluation of a beef carcass cleaning and sanitising unit. Presented at the 1979 Summer Meeting of the ASAE and CSAE. Paper No. 79-6014.

Anderson ME, Marshall RT, Stringer WC and Naumann HD, 1980. Development of design parameters for a beef carcass cleaning chamber. Presented at the 1980 Summer Meeting of the American Society Of Agricultural Engineers. Paper No. 80-6020.

Anderson ME, Marshall RT and Stringer WC, 1982. Carcass cleaning unit and containment chamber. U.S. Patent no. 4,337,548, July 16.

Anderson ME, Marshall RT, Stringer WC and Naumann HD, 1982a. Evaluation of a unique chamber for a beef carcass cleaning unit. Journal of Food Protection 45, 19-22.

Anderson ME, Marshall RT, Stringer WC and Naumann HD, 1982b. Effect of nozzle configuration and size on water absorbed by meat during cleaning. Journal of Food Protection 45, 276-278.

Anderson ME, Huff HE, Marshall RT and Stringer WC, 1982c. Inexpensive device for CIP cleaning of the inside of a beef carcass spray chamber. Journal of Food Protection 45, 643-645.

Anderson ME, Naumann HD and Cook NK, 1984. Design specifications of a red meat carcass washing and sanitising unit. Presented at the 1984 Winter Meeting of the American Society Of Agricultural Engineers. Paper No. 84-6546.

Anderson ME, Huff HE, Naumann HD, Marshall RT, Damare JM, Pratt M, and Johnston R, 1987. Evaluation of an automated beef carcass washing and sanitising system under production conditions. Journal of Food Protection 50, 562-566.

Anderson ME, Marshall RT and Dickson JS, 1991. Prediction of bacterial penetration into carcass tissue during washing. Proceedings of the 37th International Congress of Meat Science and Technology, Kulmbach, Germany, 534-537.

Anderson ME, Marshall RT, and Dickson JS, 1992. Estimating depths of bacterial penetration into post-rigor carcass tissue during washing. Journal of Food Safety 12, 191-198.

Anderson GW and Gangel MG, 1999. Method for antimicrobial treatment of animal carcasses. U.S. Patent No. 5,980,375, November 9.

Anon, 1995. Australian hot water pasteurisation (i.e. decontamination) cabinet being commercialised. Connect. No.1/2, May 1995, Publication of Australian Meat Technology Pty Ltd.

Avens JS and Miller BF, 1972. “Pasteurisation” of turkey carcasses. Poultry Science 51, 1781.

Avens JS, Albright SN, Morton AS, Prewitt BE, Kendall PA, and Sofos JN, 2002. Destruction of microorganisms on chicken carcases by steam and boiling water immersion. Food Control 13, 445–450.

Avens JS and Morton AS, 1999. Virtual eradication of psychrophilic and mesophilic bacteria on turkey carcass tails by steam and boiling water immersion. Journal of Applied Poultry Research, 8, 205-213.

Bacon RT, Belk KE, Sofos JN, Clayton RP, Reagan JO and Smith GC, 2000. Microbial populations on animal hides and beef carcasses at different stages of slaughter in plants employing multiple-sequential interventions for decontamination. Journal of Food Protection 63, 1080-1086.

Bailey C, 1971. Spray washing of lamb carcasses. Proceedings of the 17th European. Meeting of Meat Research Workers, Bristol. Paper B16, 175-181.

Bailey C, 1972. Spray washing of lamb carcasses. Institute of Meat Bulletin 75, 3-12.

Barkate ML, Acuff GR, Lucia LM and Hale DS, 1993. Hot water decontamination of beef carcasses for reduction of initial bacterial numbers. Meat Science 35, 397-401.

Berrang ME, Dickens JA and Musgrove MT, 2000. Effects of hot water application after defeathering on levels of *Campylobacter*, coliform bacteria, and *Escherichia coli* on broiler carcasses. Poultry Science 79, 1689–1693.

Bill B. and Shay B. (1995). Retail-ready meat packaging for domestic and export markets. Food Australia 48 (11), 519-522.

Bliss JN, 1975. Carcass Spraying Apparatus. British Patent 1529442.

Boerema JA and Broda DM, 2004. Microbiological Safety of Meats: *Clostridium botulinum.* In: Encyclopaedia of Meat Sciences. Eds: Jensen WK, Devine C and M. Dikeman M, Elsevier, Oxford, 786-793.

Bosilevac JM, Nou X, Barkocy-Gallagher GA, Arthur TM and Koohmaraie M, 2006. Treatments using hot water instead of lactic acid reduce levels of aerobic bacteria and Enterobacteriaceae and reduce the prevalence of *Escherichia coli* O157:H7 on pre-evisceration beef carcasses. Journal of Food Protection 69, 1808-1813.

Broda DM and Boerema JA, 2004. Microbiological Safety of Meats: *Clostridium perfringens*, Eds: Jensen WK, Devine C and M. Dikeman M, Elsevier, Oxford 2, Elsevier, Oxford, 793-798.

Bryce Jones K, 1969. Carcase cleaning. Institute of Meat Bulletin 65, 3-15.

Buhot J, Stapleton P, Green P. and Anderson P, 2000. Meat decontamination. U.S. Patent No. 6,142,861, November 7.

Buncic S. and Avery SM, 2004. Microbiological Safety of Meats: *Listeria monocytogenes*, Eds: Jensen WK, Devine C and M. Dikeman M, Elsevier, Oxford 2, Elsevier, Oxford, 804-814.

Byelashov A and Sofos JN, 2009. Strategies for on-line meat decontamination of carcasses. In: Safety of Meat and Processed Meat. Ed Toldra F, Springer Science, 149-182.

Cabedo L, Sofos JN and Smith GC, 1996. Removal of bacteria from beef tissue by spray washing after different times of exposure to fecal material. Journal of Food Protection, 59, 1284-1287.

Castelo MM, Kang D-H, Siragusa GR, Koohmaraie M and Berry ED, 2001. Evaluation of combination treatment processes for the microbial decontamination of pork trim. Journal of Food Protection, 64, 335–342.

Castillo A, Hardin MD, Acuff GR and Dickson JS, 2002. Reduction of microbial contaminants on carcasses. In: Control of Foodborne Microorganisms. Eds Juneja VK, Sofos JN, New York, Marcel Dekker, 351-381.

Castillo A, Lucia LM, Goodson KJ, Savell JW and Acuff GR, 1998. Use of hot water for beef carcass decontamination. Journal of Food Protection, 61, 19-25.

Chen H, Guan D and Hoover DG, 2005. Sensitivity of foodborne pathogens to high hydrostatic pressure. Journal of Food Protection, 69(1), 130-137.

Cho HY, Yousef AE and Sastry SK, 1999. Kinetics of inactivation of *Bacillus subtilis* spores by continuous or intermittent ohmic and conventional heating. Biotechnology and Bioengineering, 62, 368-372.

Corry JEL, James C, James SJ and Hinton M, 1995. *Salmonella*, *Campylobacter* and *Escherichia coli* O157:H7 decontamination techniques for the future. International Journal of Food Microbiology, 28, 187-196.

Corry JE, James SJ, Purnell G, Barbedo-Pinto CS, Chochois Y, Howell M and James C, 2007. Surface pasteurization of chicken carcasses using hot water. Journal of Food Engineering, 79, 913–919.

Cox NA, Mercuri AJ, Thomson JE and Gregory DW Jr, 1974. Quality of broiler carcasses as affected by hot water treatments. Poultry Science 53, 1566-1571.

Crouse JD, Anderson ME and Naumann HD, 1988. Microbial decontamination and weight of carcass beef as affected by automated washing pressure and length of time of spray. Journal of Food Protection 51, 471-474.

Cutter CN and Rivera-Betancourt M, 2000. Interventions for the reduction of *Salmonella* Typhimurium DT 104 and non-O157:H7 enterohemorrhagic *Escherichia coli* on beef surfaces. Journal of Food Protection, 63, 1326-1332.

Dahlgren D, Axelsson Olsson D, Broman T, Waldenström J, Holmberg M and Olsen B, 2003. Survival of *Campylobacter jejuni* within *Acanthamoeba polyphaga*; a possible transmission route. Arhus, Denmark 12th International Workshop on *Campylobacter*, *Helicobacter* and Related Organisms.

Davey KR, 1988. A novel cabinet for the hot water decontamination of sides of beef. Proceedings of the 34th International Congress of Meat Science and Technology, Brisbane, Australia. Part B, 642-645.

Davey KR, 1989a. Theoretical analysis of two hot water cabinet systems for decontamination of beef sides. International Journal of Food Science and Technology, 24, 291-304.

Davey KR, 1989b. Bacterial decontamination of freshly dressed beef cattle in a novel hot water cabinet. Chemeca 89, "Technology for Our Third Century", Proceedings of the 17th Australasian Chemical Engineering Conference Broadbeach, Qld., Australia, 789-796.

Davey KR and Smith MG, 1989. A laboratory evaluation of a novel hot water cabinet for the decontamination of beef sides. International Journal of Food Science and Technology, 24, 305-316.

Davey KR 1990a. A model for the hot water decontamination of sides of beef in a novel cabinet based on laboratory data. International Journal of Food Science and Technology 25, 88-97.

Davey KR, 1990b. Decontamination of meat. U.S. Patent no. 4,965,911, October 30.

Dawson LE, Mallmann WL, Bigbee DG, Walker R, Zabik ME, 1963 Influence of surface pasteurisation and chlortetracycline on bacterial incidence on fryers. Food Technology 17, 100-104.

Delmore Jr RJ, Sofos JN, Schmidt GR, Belk KE, Lloyd WR and Smith GC, 2000. Interventions to reduce microbiological contamination of beef variety meats. Journal of Food Protection, 63, 44-50.

Dincer AH and Baysal T, 2004. Decontamination techniques of pathogen bacteria in meat and poultry. Critical Reviews in Microbiology 30, 197-204.

Donnison AM and Ross CM, 2003. Is *Campylobacter* and faecal microbial contamination a problem in rural streams? In Environmental Management Using Soil-plant Systems. Ed Currie LD, Stewart RB and Andersen CWN. Fertilizer and Lime Research Centre Occasional Report 16, Palmerston North: Massey University, 234–241.

Dorsa WJ, 1997. New and established carcass decontamination procedures commonly used in the beef-processing industry. Journal of Food Protection, 60, 1146-1151.

Dorsa WJ, Cutter CN and Siragusa GR, 1997. Effects of steam-vacuuming and hot water spray wash on the microflora of refrigerated beef carcass surface tissue inoculated with *Escherichia coli* O157:H7, *Listeria innocua*, and *Clostridium sporogenes*. Journal of Food Protection, 60, 114-119.

Duffy G, Butler F, Cummins E, O'Brien S, Nally P, Carney E, Henchion M, Mahon D, Cowan C, 2006. *E. coli* O157:H7 in beefburgers produced in the republic of Ireland: a quantitative microbial risk assessment. Ireland, Tgc.

EFSA Panel on Biological Hazards (BIOHAZ), 2006. Joint AFC/BIOHAZ guidance document on the safety and the efficacy of substances for the removal of microbial surface contamination of foods of animal origin. The EFSA Journal 388: 1-19.

EFSA Panel on Biological Hazards (BIOHAZ), 2007. Scientific Opinion of the Panel on Biological Hazards on a request from EFSA on monitoring of verotoxigenic *Escherichia coli* (VTEC) and identification of human pathogenic VTEC types. The EFSA Journal 579, 1-61.

EFSA Panel on Biological Hazards (BIOHAZ), 2009. Scientific Opinion of the Panel on Biological Hazards on a request from European Commission on the use and mode of action of bacteriophages in food production. The EFSA Journal (2009) 1076, 1-26

EFSA Panel on Biological Hazards (BIOHAZ),2010. Guidance on Revision of the joint AFC/BIOHAZ guidance document on the submission of data for the evaluation of the safety and efficacy of substances for the removal of microbial surface contamination of foods of animal origin intended for human consumption. EFSA Journal 2010; 8(4):1544.

Eggenberger-Solorzano L, Niebuhr SE, Acuff GR and Dickson JS, 2002. Hot water and organic acid interventions to control microbiological contamination on hog carcasses during processing. Journal of Food Protection, 65, 1248–1252.

Ellebracht EA, Castillo A, Lucia LM, Miller RK and Acuff GR, 1999. Reduction of pathogens using hot water and lactic acid on beef trimmings. Journal of Food Science, 64, 1094-1099.

Feirtag JM and Pullen MM, 2003. A novel intervention for the reduction of bacteria on beef carcasses. Food Protection Trends 23: 558-562.

Gill CO and Badoni M, 1997a. The hygienic and organoleptic qualities of ground beef prepared from manufacturing beef pasteurized by immersion in hot water. Meat Science, 46, 67-75.

Gill CO and Badoni M, 1997b. The effects of hot water pasteurizing treatments on the appearances of pork and beef. Meat Science, 46, 77-87.

Gill CO and Badoni M, 2002. Microbiological and organoleptic qualities of vacuum-packaged ground beef prepared from pasteurized manufacturing beef. International Journal of Food Microbiology, 74, 111–118.

Gill CO and Badoni M, 2003. Effects of storage under a modified atmosphere on the microbiological and organoleptic qualities of ground beef prepared from pasteurized manufacturing beef. International Journal of Food Science and Technology, 38, 233-240.

Gill CO and Bryant J, 2000. The effects on product of a hot water pasteurizing treatment applied routinely in a commercial beef carcass dressing process. Food Microbiology, 17, 495-504.

Gill CO and Jones T, 1998. Control of the contamination of pig carcasses by *Escherichia coli* from their mouths. International Journal of Food Microbiology 44, 43-48.

Gill CO, Bedard D and Jones T, 1997. The decontaminating performance of a commercial apparatus for pasteurizing polished pig carcasses. Food Microbiology 14:1, 71-79.

Gill CO, Bedard D and Jones T, 1997. The decontaminating performance of a commercial apparatus for pasteurizing polished pig carcasses. Food Microbiology, 14, 71–79.

Gill CO, Bryant J and Badoni M, 2001. Effects of hot water pasteurizing treatments on the microbiological condition of manufacturing beef used for hamburger patty manufacture. International Journal of Food Microbiology, 63, 243-256.

Gill CO, Bryant J and Bedard D, 1999. The effects of hot water pasteurizing treatments on the appearances and microbiological conditions of beef carcass sides. Food Microbiology, 16, 281-289.

Gill CO, Jones T and Badoni M, 1998. The effects of hot water pasteurizing treatments on the microbiological conditions and appearances of pig and sheep carcasses. Food Research International, 43, 273-278.

Gill CO, McGinnis DS, Bryant J and Chabot B, 1995. Decontamination of commercial, polished pig carcasses with hot water. Food Microbiology 12, 143-149.

Göksoy, E. O., James, C., Corry, J. E. L. and James, S. J. (2001). The effect of hot water immersions on the appearance and microbiological quality of skin-on chicken breast pieces. International Journal of Food Science and Technology. 36:1, 61-69.

Gorman BM, Kochevar SL, Sofos JN, Morgan JB, Schmidt GR and Smith GC, 1997. Changes on beef adipose tissue following decontamination with chemical solutions or water of 35°C or 74°C. Journal of Muscle Foods, 8, 185-197.

Gorman BM, Morgan JB, Sofos JN and Smith GC, 1995a. Microbiological and visual effects of trimming and/or spray washing for removal of fecal material from beef. Journal of Food Protection, 58, 984-989.

Gorman BM, Sofos JN, Morgan JB, Schmidt GR and Smith GC, 1995b. Evaluation of hand-trimming, various sanitizing agents and hot water spray-washing as decontamination interventions of beef brisket adipose tissue. Journal of Food Protection, 58, 899-907.

Gould GW, 2006. History of science – spores. Journal Applied Microbiology 101, 507-513.

Gracey JF, Collins DS and Huey RJ, 1999. Meat hygiene. Elsevier Health Sciences, 758 pp.

Graham A, Cain BP and Eustace IJ, 1978. An enclosed hot water spray cabinet for improved hygiene of carcass meat. CSIRO Meat Research Report No. 11/78.

Graham A, 1979. A hot shower for clean carcasses. Australian Refrigeration Air Conditioning And Heating 33, 33-34.

Graves Delmore LR, Sofos JN, Reagan JO and Smith GC, 1997. Hot-water rinsing and trimming/washing of beef carcasses to reduce physical and microbiological contamination. Journal of Food Science, 62, 373-376.

Graves Delmore LR, Sofos JN, Schmidt GR and Smith GC, 1998. Decontamination of inoculated beef with sequential spraying treatments. J. Food Sci. 63:890- 893.

Griffis CL and Osaili TM, 2009 Control of thermal meat processing. In: Todra F. (Ed.) Safety of Meat and Processed Meat. Springer, New York, 229-253.

Griffiths MW, 2009. *Bacillus cereus*. In Pathogens and Toxins, Challenges and Interventions. Ed: Juneja VK and Sofos JN. ASM Press, Washington, DC, 1-19.

Guerrero-Legarreta I, 2004. Canning. In Encyclopaedia of Meat Sciences, Eds Jensen WK, Devine C and Dikeman M, Elsevier, Oxford, 139-144.

Harben GS Jr, 1989. Fowl scalding apparatus and method. U.S. Patent No. 4,868,950, September 26.

Hauschild AHW, 1989. *Clostridium botulinum*. In Foodborne Bacterial Pathogens, Ed Doyle MP, Marcel Dekker Inc., New York, NY, USA, 111-189.

Heller CE, Scanga JA, Sofos JN, Belk KE, Warren-Serna W, Bellinger GR, Bacon RT, Rosman ML, and Smith GC, 2007. Decontamination of beef subprimal cuts intended for blade tenderization or moisture enhancement. Journal of Food Protection, 70, 1174-1180.

Hood SK, and Zottola EA, 1997. Adherence to stainless steel by foodborne microorganisms during growth in model food systems. International Journal of Food Microbiology 22, 145-53.

Hudson JA, 2004. *Staphylococcus aureus*. In: Encyclopaedia of Meat Sciences, Eds Jensen WK, Devine C and Dikeman M, Elsevier, Oxford, 820-825

Huffman RD, 2002. Current and future technologies for the decontamination of carcasses and fresh meat. Meat Science 62, 285–294.

Hugas M and Tsigarida E, 2008. Pros and cons of carcass decontamination: The role of the European Food Safety Authority. Meat Science, 78, 43-52.

ICMSF (International Commission on Microbiological Specifications for Foods). 1996. Microorganisms in Foods, In Microbiological Specifications of Food Pathogens, Blackie academic, Professional, London.

ICMSF (International Commission on Microbiological Specifications for Foods). 1998. Microorganisms in Foods, Microbial Ecology of Food Commodities, Blackie academic, Professional, London.

Ikeda JS, Samelis J, Kendall PA, Smith GC and Sofos JN, 2003. Acid adaptation does not promote survival or growth of *Listeria monocytogenes* on fresh beef following acid and nonacid decontamination treatments. Journal of Food Protection 66, 985-992.

James C, James SJ, Hannay N, Purnell G, Barbedo-Pinto C, Yaman H, Araujo M, Gonzalez ML, Calvo J, Howell M and Corry JEL, 2007. Decontamination of poultry carcasses using steam or hot water in combination with rapid cooling, chilling or freezing of carcass surfaces. International Journal of Food Microbiology, 114, 195–203.

James C, Thornton JA, Ketteringham L and James SJ, 2000. Effect of steam condensation, hot water or chlorinated hot water immersion on bacterial numbers and quality of lamb carcasses. Journal of Food Engineering, 43, 219-225.

Johnson EA, 2007. *Clostridium botulinum*. In Pathogens and Toxins, Challenges and Interventions. Eds Juneja VK and Sofos JN. ASM Press, Washington, DC, 401-421.

Juneja VK, Novak JS and Labbe RJ, 2009. *Clostridium perfringens*. In Pathogens and Toxins, Challenges and Interventions. Eds Juneja VK and Sofos JN. ASM Press, Washington, DC, 53-70.

Kalchayanand N, Arthur TM, Bosilevac JM, Brichta-Harhay DM, Guerini MN, Wheeler TL and Koohmaraie M, 2008. Evaluation of various antimicrobial interventions for the reduction of *Escherichia coli* O157:H7 on bovine heads during processing. Journal of Food Protection, 71, 621-624.

Kang DH, Koohmaraie M, Dorsa WJ and Siragusa GR, 2001. Development of a multiple-step process for the microbial decontamination of beef trim. Journal of Food Protection, 64, 63-71.

Keener KM, Bashor MP, Curtis PA, Sheldon BW and Kathariou S, 2004. Comprehensive review of *Campylobacter* and poultry processing. Journal of Food Science, 3, 105-116.

Kelly CA, Dempster JF and McLoughlin AJ, 1981. The effect of temperature, pressure and chlorine concentration of spray washing water on numbers of bacteria on lamb carcasses. Journal of Applied Bacteriology. 51, 415-424.

Kelly CA, Lynch JF and McLoughlin AJ, 1982. The effect of spray washing on the development of bacterial numbers and storage life of lamb carcasses. Journal of Applied Bacteriology 53, 335-341.

Kiermeier A, Bobbitt J, Vanderlinde P, Higgs G, Pointon A and Sumner J, 2006. Use of routine beef carcase *Escherichia coli* monitoring data to investigate the relationship between hygiene status of incoming stock and processing efficacy. International Journal of Food Microbiology, 111, 263–269.

Kochevar SL, Sofos JN, LeValley SB and Smith GC, 1997. Effect of water temperature, pressure and chemical solution on removal of fecal material and bacteria from lamb adipose tissue by spray-washing. Meat Science, 45, 377-388.

Koutsoumanis KP, Ashton LV, Geornaras I, Belk KE, Scanga JA, Kendall PA, Smith GC and Sofos JN, 2004. Effect of single sequential hot water and lactic acid decontamination treatments on the survival and growth of *Listeria monocytogenes* and spoilage microflora during aerobic storage of fresh beef at 4, 10 and 25°C. Journal of Food Protection, 67, 2703-2711.

Kramer JM and Gilbert RJ, 1989. *Bacillus cereus* and Other *Bacillus* Species. In Foodborne Bacterial Pathogens, Ed Doyle MP, Marcel Dekker, Inc., New York, NY, USA, 21-70.

Labbe R, 1989. *Clostridium perfringens*. In Foodborne Bacterial Pathogens, Ed Doyle MP, Marcel Dekker, Inc., New York, NY, USA, 191-234.

Latha C, Sherikar AT, Waskar VS, Dubal ZB and Ahmed SN, 2009. Sanitizing effect of salts on experimentally inoculated organisms on pork carcasses. Meat Science, 83, 796–799.

Ledesma AMR, Riemann HP and Farver TB, 1996. Short-time treatment with alkali and/or hot water to remove common pathogenic and spoilage bacteria from chicken wing skin. Journal of Food Protection 59, 746-750.

Li Y, Yang H and Swem BL, 2002. Effect of high-temperature inside-outside spray on survival of *Campylobacter jejuni* attached to pre-chill chicken carcasses. Poultry Science, 81, 1371–1377.

Loretz M, Stephan R and Zweifel C, 2010. Antimicrobial activity of decontamination treatments for poultry carcasses: A literature survey. Food Control, 21, 791–804.

Lou YQ and Yousef AE, 1997. Adaptation to sublethal environmental stresses protects *Listeria monocytogenes* against lethal preservation factors. Applied and Environmental Microbiology, 63, 1252-1255.

Madigan M, Martinko J, Parker J, 1997. Host-parasite relationships. In Brock biology of microorganisms, Prentice-Hall, Inc., Upper Saddle River, NJ, 789-817.

McGinnis DS, Gill CO, Chabot BP and Popovich WJ, 1997. Pasteurization of carcasses with directed sheets of heated water. U.S. Patent No. 5,651,730, July 29.

Mekalanos JJ, 1992. Environmental signals controlling expression of virulence determinants in bacteria. Journal of Bacteriology, 174, 1-7.

Mills J, 2004. Microbiological Safety of Meats: *Yersinia enterocolitica*. In Encyclopaedia of Meat Sciences, Eds Jensen WK, Devine C and Dikeman M, Elsevier, Oxford, 814-820.

Morgan AI, Goldberg N, Radewonuk ER, Scullen OJ, 1996a. Surface pasteurisation of raw poultry meat by steam. Lebensmittel -Wissenschaft und -Technologie 29, 447-451.

Morgan AI, Radewonuk ER and Scullen OJ, 1996b. Ultra high temperature, ultra short time surface pasteurisation of meat. Journal of Food Science 61, 1216-1218.

Morrison GJ and Fleet GH, 1985. Reductions of *Salmonella* on chicken carcasses by immersion treatments. Journal of Food Protection 48, 939-943.

Muraca P, Stout JE and Yu VL, 1987. Comparative assessment of chlorine, heat, ozone, and UV light for killing *Legionella pneumophila* within a model plumbing system. Applied Environmental Microbiology, 53, 447-53.

Nørrung B, Andersen JK, Buncic S, 2009. Main Concerns of Pathogenic Microorganisms in Meat Food Microbiology and Food Safety, 3-29.

Nørrung B and Buncic S, 2008 Microbial safety of meat in the European Union. Meat Science 78, 1-2, 14-24.

Northcutt JK, Smith DP, Musgrove MT, Ingram KD and Hinton A Jr, 2005. Microbiological impact of spray washing broiler carcasses using different chlorine concentrations and water temperatures. Poultry Science, 84, 1648–1652.

Novak JS, Yuan JTC, 2004. The fate of *Clostridium perfringens* spores exposed to ozone and/or mild heat pretreatment on beef surfaces followed by modified atmosphere packaging. Food Microbiology 21: 667-673.

O'Driscoll B, Gahan CGM, Hill C, 1996. Adaptive acid tolerance response in *Listeria monocytogenes*: Isolation of an acid-tolerant mutant which demonstrates increased virulence. Applied Environmental Microbiolology, 62, 1693-1698.

Özdemir H, Koluman A and Yildirim Y, 2006a. Effects of acidified sodium chlorite, cetylpyridinium chloride and hot water on populations of *Listeria monocytogenes* and *Staphylococcus aureus* on beef. Letters in Applied Microbiology, 43, 168-173.

Özdemir H, Yildirim Y, Küplülü O, Koluman A, Göncüoğlu M and Gökhan I, 2006b. Effects of lactic acid and hot water treatments on *Salmonella* Typhimurium and *Listeria monocytogenes* on beef. Food Control, 17, 299-303.

Peck MW, 2009. *Clostridium botulinum*. In Pathogens and Toxins, Challenges and Interventions. Eds Juneja VK and Sofos JN. ASM Press, Washington, DC. 31-52.

Phebus RK, Nutsch AL, Schafer DE, Wilson RC, Riemann MJ, Leising JD, Kastner CL, Wolf JR and Prasai RK, 1997. Comparison of steam pasteurization and other methods of reduction of pathogens on surfaces of freshly slaughtered beef. Journal of Food Protection 60, 476–484.

Pickett LD and Miller BF, 1966. The effect of high temperature immersion on the microbial population of whole turkey carcasses. Poultry Science. 45, 1116.

Pordesimo LO, Wilkerson EG, Womac AR and Cutter CN, 2002. Process engineering variables in the spray washing of meat and produce. Journal of Food Protection 65:1, 222-237.

Powell VH and Cain BP, 1987. A hot water decontamination system for beef sides. CSIRO Food Research Quarterly 47:4, 79-84.

Purnell G, Mattick K and Humphrey T, 2004. The use of 'hot wash' treatments to reduce the number of pathogenic and spoilage bacteria on raw retail poultry. Journal of Food Engineering 62:1, 29-36.

Reagan JO, Acuff GR, Buege DR, Buyck MJ, Dickson JS, Kastner CL, Marsden JL, Morgan JB, Nickelson R, Smith GC and Sofos JN, 1996. Trimming and washing of beef carcasses as a method of improving the microbiological quality of meat. Journal of Food Protection, 59, 751-756.

Reichert N, 1979. Aspects of meat product manufacture and preservation. Fleischerei 30(5) 416-418.

Rivas L, Dykes GA and Fegan N, 2006. Attachment of shiga toxigenic *Escherichia coli* to beef muscle and adipose tissue. Journal of Food Protection 69(5): 999-1006.

Rodriguez-Palacios A, Staempfli HR, Duffield T, Peregrine AS, Trotz-Williams LA, Arroyo LG, Brazier JS and Weese JS, 2006. *Clostridium difficile* PCR ribotypes in calves, Canada. Emerging Infectious Disease 12, 1730–1736.

Rodriguez-Palacios A, Reid-Smith RJ, Staempfli HR and Scott Weese J, in press. *Clostridium difficile* survives minimal temperature recommended for cooking ground meats. Anaerobe.

Samelis J and Sofos JN, 2003. Strategies to control stress-adapted pathogens. In: Microbial Stress Adaptation and Food Safety, Eds Yousef AE, Juneja VK. Boca Raton: CRC Press, 303-351.

Schaefer DL, Leising JD and Riemann MJ, 2006. Surface treatment of animal tissue. U.S. Patent No. 7,108,882 B2, September 19.

Schulze-Roebbecke R, Rodder M and Exner M, 1987. Multiplication and killing temperatures of naturally occurring legionellas. Zentralbl Bakteriol Mikrobiol Hyg. 184(6), 495-500.

SCVPH (Scientific Committee On Veterinary Measures Relating To Public Health), 1998. Report on the benefits and limitations of antimicrobial treatments for poultry carcasses. 30 October 1998.

SCVPH (Scientific Committee On Veterinary Measures Relating To Public Health), 2003. Opinion on the evaluation of antimicrobial treatments for poultry carcasses. Opinion issued on 14-15 April 2003.

Setlow P and Johnson EA, 2007. Spores and their significance. In Food Microbiology, Fundamentals and Frontiers. Eds Doyle MP and Beuchat LR. ASM Press, Washington, DC. 35-67.

Shadbolt C, Ross T and McMeekin TA, 2001. Differentiation of the effects of lethal pH and water activity: food safety implications. Letters in Applied Microbiology, 32, 99-102.

Sheridan JJ, 1982. Problems associated with commercial lamb washing in Ireland. Meat Science 6, 211-219.

Sheridan JJ, 2004. Decontamination. In Encyclopaedia of Meat Sciences, Eds Jensen WK, Devine C and Dikeman M, Elsevier, Oxford, 1-25.

Sinhamahapatra M, Biswas S, Das AK and Bhattacharyya D, 2004. Comparative study of different surface decontaminants on chicken quality. British Poultry Science, 45, 624–630.

Smith MG, 1992. Destruction of bacteria on fresh meat by hot water. Epidemiology and Infection, 109, 491-496.

Smith MG and Davey KR, 1990. Destruction of *Escherichia coli* on sides of beef by a hot water decontamination process. Food Australia, 42, 195–198.

Smith MG and Graham A, 1978. Destruction of *Escherichia coli* and salmonellae on mutton carcasses by treatment with hot water. Meat Science, 2, 119-128.

Smulders FJ and Greer GG, 1998. Integrating microbial decontamination with organic acid in HACCP programmes from muscle foods: Prospects and controversies. Journal of Food Microbiology, 44: 149.1 69.

Sofos JN, 2005. Improving the safety of fresh meat. Boca Raton, FL: CRC Press.

Sofos JN, 1989. Detection of injured sporeforming bacteria from foods. In Injured Index and Pathogenic Bacteria: Occurrence and Detection in Foods, Water and Feeds. Ed B. Ray. CRC Press, Boca Raton, FL. 147-177.

Sofos JN and Smith GC, 1998. Non-acid meat decontamination technologies: model studies and commercial applications. International Journal of Food Microbiology, 44, 171-188.

Stivarius MR, Pohlman FW, McElyea KS, Waldroup AL, 2002. Effects of hot water and lactic acid treatment of beef trimmings prior to grinding on microbial, instrumental colour and sensory properties of ground beef during display. Meat Science, 60, 327-334.

Stout JE, Best MG and Yu VL, 1986. Susceptibility of members of the family Legionellaceae to thermal stress: implications for heat eradication methods in water distribution systems. Applied Environmental Microbiology 52(2): 396–399.

Stumbo CR, 1973 "Thermobacteriology in Food Processing", Academic Press, London.

Karlsson S, Dupuy B, Mukherjee K, Norin E, Burman LG and Åkerlund T, 2003. Expression of *Clostridium difficile* Toxins A and B and Their Sigma Factor TcdD Is Controlled by Temperature. Infection and Immunity, 71(4), 1784-1793.

Teilmann J, 2002. System for treatment of carcass halves with hot water on a slaughter line. International Publication number WO 02/39821 A1.

Teotia JS and Miller BF, 1972. Destruction of S*almonella* on turkey carcass skin with chemicals and hot water. Poultry Science 51, p1878.

Thomson JE, Bailey JS and Cox NA, 1979. Phosphate and heat treatments to control *Salmonella* and reduce spoilage and rancidity on broiler carcasses. Poultry Science. 58, 139-143.

Thomson JE, Cox NA, Whitehead WK, Mercuri AJ, 1974. Effect of hot spray washing on broiler carcass quality. Poultry Science. 53, 946-952.

Van Netten P, Mossel DAA and Huis In JHJ, (1997). Microbial changes on freshly slaughtered pork carcasses due to "hot" lactic acid decontamination. Journal of Food Safety, 17, 89-111.

Varnam AH and Sutherland JP, 1995. Meat and Meat products. Chapman, Hall. Food Products Series, Vol 3.

Warriner K, Eveleigh K, Goodman J, Betts G, Gonzales M and Waites WM, 2001. Attachment of Bacteria to Beef from Steam-Pasteurized Carcasses. Journal of Food Protection, 4, 493-497(5).

WHO/FAO, 2002. Risk Assessments of *Salmonella* in Eggs and Broiler Chickens. Food and Agriculture Organization of the United Nations and the World Health Organization.

Whyte P, McGill K and Collins JD, 2003. An assessment of steam pasteurization and hot water immersion treatments for the microbiological decontamination of broiler carcasses. Food Microbiology, 20, 111-117.

Whyte RJ and Wong TL, 2004. *Bacillus cereus*. In Encyclopaedia of Meat Sciences, Eds Jensen WK, Devine C and Dikeman M, Elsevier, Oxford, 825-830.

Willshaw GA, Cheasty T and Smith HR, 2000. *Escherichia coli*. In The Microbiological Safety and Quality of Food, Eds Lund BM, Baird-Parker TC and Gould GW, Aspen Publishers, Gaithersburg, MD, 1136–1177.

Zuniga AG, Anderson EM, Marshall RT and Iannotti EL, 1991. A model system for studying the penetration of microorganisms into meat. Journal of Food Protection 54, 256-258.

APPENDICES

A. TECHNICAL SPECIFICATION OF DIFFERENT TECHNIQUES USED IN TRIALS FOR THE EVALUATION OF CARCASS DECONTAMINATION TREATMENTS THROUGH THE USE OF HOT WATER SYSTEMS

SPRAYING

1. Hot water treatments (>70°C) reduce bacterial counts by 2 to 3 log CFU/cm^2.
2. Water pressures ranging from 1379 to 2070 kPa are most effective in reducing microbial contamination.
3. A water flow rate of 7.5 lmin^{-1} is recommended for effective microbial removal.
4. Efficacy of spray treatment can be improved by the use of more than one spray treatment or by the combination with treatment followed by the application of sanitising agents.

MANUAL SPRAY

As example, the optimal specifications of the trials by Bailey (1971) are reported:

- Spray type: Fan jet system
- Dynamic line pressure: 690 kNm^{-2}
- Surface impact: 1.035 kNm^{-2}
- Water supply temperature: 90°C
- Surface spray temperature: 60°C
- Water flow rate: 8.5 litres/min

SPRAY CABINETS

OPTIMAL SPECIFICATIONS OF THE COMMERCIAL CAPER SYSTEM UNIT (ANDERSON ET AL. 1984)

- Spray type: two oscillating spray bars on a vertical axis of 45°
- Dynamic line pressure: 172 to 4134 kNm^{-2}
- Water supply temperature: 16°C
- Water flow rate: 0 to 378 litres/min for treatment unit

  0 to 189 litres/min for sanitising unit

RELATIONSHIP BETWEEN LINE PRESSURE AND DYE PENETRATION DURING INVESTIGATIONS INTO BACTERIAL PENETRATION INTO MEAT WITH TREATMENT USING BLUE LAKE DYE (DE ZUNIGA ET AL. 1991)

$D = 0.471 + 0.000154 (X)$

where D = penetration (mm) and X = line pressure (kNm^{-2}).

Further predictive equations for bacterial penetration were developed by Anderson et al.(1991; 1992b) relating to different types of meat tissue:

Exterior lean:

$D = 0.262 + 0.014 (X) - 6.525 \times 10^{-5} (X^2) + 1.040 \times 10^{-7} (X^3) - 5.322 \times 10^{-11} (X^4)$

Exterior fat:

$D = 0.25 + 0.015 (X) - 7.156 \times 10^{-5} (X^2) + 1.224 \times 10^{-7} (X^3) - 6.695 \times 10^{-11} (X^4)$

Cut surface:

$D = 0.25 + 0.017 (X) - 6.015 \times 10^{-5} (X^2)$

Interior body cavity:

$D = 0.25 + 0.009 (X) - 4.609 \times 10^{-5} (X^2) + 8.184 \times 10^{-8} (X^3) - 4.517 \times 10^{-11} (X^9)$

SPECIFICATIONS OF FULLY ENCLOSED SPRAY CABINET SYSTEM (GRAHAM et al. 1978)

- Spray type: 32 water jets mounted on eight distribution pipes.
- Dynamic line pressure: 200 kNm^{-2}
- Water supply temperature: 90°C
- Surface spray temperature: 80°C
- Water flow rate: 270 litres/min
- Throughput: 300 carcasses/h

SPECIFICATIONS OF ADAPTED SPRAY CABINET SYSTEM (POWELL AND CAIN, 1987)

- Spray type: two banks of eight nozzles.
- Dynamic line pressure: 300 kNm^{-2}
- Water supply temperature: 80-85°C
- Surface spray temperature: 73-78°C
- Water flow rate: 222 litres/min
- Throughput: 135 carcasses/h

B. EFFICACY OF HOT POTABLE WATER IN REDUCING BACTERIA COUNTS ON THE SURFACE OF MEAT CARCASSES IN DIFFERENT SPECIES

Table 1: Efficacy of hot potable water in reducing bacteria counts on the surface of beef carcasses and parts

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (s)	Sampling point	Reduction (log CFU)	Reference
Aerobic bacteria	SP	Carcass	Natural	95	12	Before final carcass wash	1.3 cm^{-2}	Barkate et al. (1993)
	SP	Carcass	Natural	95	12	After final carcass wash	0.8 cm^{-2}	Barkate et al. (1993)
	SP	Brisket adipose tissue	Artificial	74	12	During slaughter	1.7-2.2 cm^{-2}	Gorman et al. (1995a)
	SP	Short plates	Artificial	72	12	After slaughter	2.0 cm^{-2}	Dorsa et al. (1996)
	SP	Short plates	Natural	72	12	After slaughter	0.3 cm^{-2}	Dorsa et al. (1996)
	SP	Carcass	Artificial	74-87.8	11-18	During slaughter	2.0 cm^{-2}	Reagan et al. (1996)
	SP	Short plates	Artificial	74 followed by 30	15/wash temp	After slaughter	2.1 cm^{-2}	Dorsa et al. (1997)
	IM	Meat pieces[b]	Natural	85	15 60	Packing plant	1.1 g^{-1} 1.3 g^{-1}	Gill and Badoni (1997a)
	SP	Chuck adipose tissue	Artificial	74	12	During slaughter	2.8 cm^{-2}	Gorman et al. (1997)
	SP	Carcass	Artificial	>77	2.5 8	During slaughter	0.2-0.3 cm^{-2} 0.0-0.2 cm^{-2}	Graves Delmore et al. (1997)
	SP	Carcass surface regions	Artificial	95	5	After slaughter	2.9 cm^{-2}	Castillo et al. (1998)
	SP	Brisket adipose tissue	Artificial	80	5.6	During slaughter	0.2 cm^{-2}	Graves Delmore et al. (1998)

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (s)	Sampling point	Reduction (log CFU)	Reference
	IM	Trimmings	Artificial	95	3	Processing facilities (before grinding)	0.3 g^{-1}	Ellebracht et al. (1999)
	SP	Carcass	Natural	85	10 and 15	Dressing	1.5 cm^{-2}	Gill et al. (1999)
	IM	Variety meats	Natural	80	10	Packing plant (packaging)	0.5-1.4 g^{-1}	Delmore, Jr. et al. (2000)
	SP	Variety meats	Natural	78-80	10	Packing plant (packaging)	0.1-1.5 g^{-1}	Delmore, Jr. et al. (2000)
	IM	Meat pieces[b]	Natural	85	60	Packing plant	>1.0 g^{-1}	Gill and Badoni (2002)
	IM	Trimmings[b]	Artificial	82	180	NA[c]	0.0 g^{-1}	Stivarius et al. (2002)
	IM	Meat pieces[b]	Natural	85	60	Packing plant	>1.0 g^{-1}	Gill and Badoni (2003)
	SP	Carcass	Natural	74	5.5	Before evisceration	2.7 $(10\ cm)^{-2}$	Bosilevac et al. (2006)
Aeromonas hydrophila	SP	Brisket	Artificial	80	10 20	After slaughter	3.2 cm^{-2} 3.3 cm^{-2}	Smith (1992)
Brochothrix thermosphacta	IM	Meat pieces[b]	Natural	85	60	Packing plant	>1.0 g^{-1}	Gill and Badoni (2002)
Clostridium sporogenes	SP	Short plates	Artificial	74 followed by 30	15/wash temp	After slaughter	2.7 cm^{-2}	Dorsa et al. (1997)
Coliforms	SP	Short plates	Artificial	72	12	After slaughter	2.7 cm^{-2}	Dorsa et al. (1996)
	SP	Carcass	Artificial	>77	2.5 8	During slaughter	1.4-1.6 cm^{-2} 1.3-1.8 cm^{-2}	Graves Delmore et al. (1997)
	SP	Brisket adipose tissue	Artificial	80	5.6	During slaughter	0.9 cm^{-2}	Graves Delmore et al. (1998)
	SP	Carcass	Natural	85	10 and 15	Dressing	2.0 cm^{-2}	Gill et al. (1999)
	IM	Variety meats	Natural	80	10	Packing plant (packaging)	0.7-1.6 g^{-1}	Delmore, Jr. et al. (2000)

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (s)	Sampling point	Reduction (log CFU)	Reference
	SP	Variety meats	Natural	78-80	10	Packing plant (packaging)	0.0-1.6 g^{-1}	Delmore, Jr. et al. (2000)
	SP	Carcass	Natural	85	8-12	Dressing	1.0->2.0 cm^{-2}	Gill and Bryant (2000)
	SP	Trim	Artificial	65 71 76 82	3 passes under spray bar	NA[c]	1.7 cm^{-2} 1.7 cm^{-2} 2.3 cm^{-2} 2.8 cm^{-2}	Kang et al. (2001)
	IM	Trimmings[b]	Artificial	82	180	NA[c]	0.1 g^{-1}	Stivarius et al. (2002)
Escherichia coli	IM	Carcass	Artificial	80	10	After slaughter and chilling	2.2-3.0 cm^{-2}	Smith and Graham (1978)
	SP	Carcass	Natural	83.5	20	After slaughter	3.0 cm^{-2}	Smith and Davey (1990)
	SP	Brisket	Artificial	80	10 20	After slaughter	3.1 cm^{-2} 3.3 cm^{-2}	Smith (1992)
	SP	Brisket adipose tissue	Artificial	74	12	After exsanguination	2.6-4.2 cm^{-2}	Cabedo et al. (1996)
	SP	Short plates	Artificial	72	12	After slaughter	2.7 cm^{-2}	Dorsa et al. (1996)
	SP	Carcass	Artificial	74-87.8	11-18	During slaughter	1.7 cm^{-2}	Reagan et al. (1996)
	SP	Carcass	Natural	85	10 and 15	Dressing	2.0 cm^{-2}	Gill et al. (1999)
	IM	Variety meats	Natural	80	10	Packing plant (packaging)	0.0-1.6 g^{-1}	Delmore, Jr. et al. (2000)
	SP	Variety meats	Natural	78-80	10	Packing plant (packaging)	0.0-2.0 g^{-1}	Delmore, Jr. et al. (2000)
	IM	Trimmings[b]	Artificial	82	180	NA[c]	0.0 g^{-1}	Stivarius et al. (2002)

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (s)	Sampling point	Reduction (log CFU)	Reference
***E. coli* O157:H7**	SP	Short plates	Artificial	74 followed by 30	15/wash temp	After slaughter	2.6 cm^{-2}	Dorsa et al. (1997)
	IM	Trimmings	Artificial	95	3	Processing facilities (before grinding)	0.5 g^{-1}	Ellebracht et al. (1999)
	SP	Carcass surface regions	Artificial	95	5	After slaughter	3.7 cm^{-2}	Castillo et al. (1998)
	SP	Beef surfaces	Artificial	72	15	After exsanguination	0.8-1.9 cm^{-2}	Cutter and Rivera-Betancourt (2000)
	SP	Subprimal cuts	Artificial	82	20	Processing plants	1.0 cm^{-2}	Heller et al. (2007)
	SP	Heads	Artificial	74	26	Processing plant	1.7 cm^{-2}	Kalchayanand et al. (2008)
Enterobacteriaceae	SP	Carcass	Natural	74	5.5	Before evisceration	2.7 $(10\ cm)^{-2}$	Bosilevac et al. (2006)
	IM	Meat pieces[b]	Natural	85	60	Packing plant	<1.0 g^{-1}	Gill and Badoni (2002)
Enterohemorrhagic *E. coli* (non-O157:H7)	SP	Beef surfaces	Artificial	72	15	After exsanguination	1.8-2.2 cm^{-2}	Cutter and Rivera-Betancourt (2000)
Enteropathogenic *E. coli*	SP	Brisket	Artificial	80	10 20	After slaughter	2.6 cm^{-2} 3.0 cm^{-2}	Smith (1992)
Lactic acid bacteria	IM	Meat pieces[b]	Natural	85	60	Packing plant	<1.0 g^{-1}	Gill and Badoni (2002)
	IM	Meat pieces[b]	Natural	85	60	Packing plant	0.5 g^{-1}	Gill and Badoni (2003)
Listeria monocytogenes	SP	Brisket	Artificial	80	10 20	After slaughter	2.8 cm^{-2} 2.9 cm^{-2}	Smith (1992)
	IM	Leg (top-round)	Artificial	75	30	After slaughter	0.8 cm^{-2}	Koutsoumanis et al. (2004)
	IM	Muscle	Artificial	93	15	Retail	2.1 g^{-1}	Özdemir et al. (2006a)
	IM	Muscle	Artificial	82	15	Retail	0.1 g^{-1}	Özdemir et al. (2006b)

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (s)	Sampling point	Reduction (log CFU)	Reference
***Pseudomonas* spp.**	SP	Brisket	Artificial	80	10 20	After slaughter	3.0 cm^{-2} 3.4 cm^{-2}	Smith (1992)
	IM	Meat pieces[b]	Natural	85	60	Packing plant	>1.0 g^{-1}	Gill and Badoni (2002)
	IM	Meat pieces[b]	Natural	85	60	Packing plant	>1.0 g^{-1}	Gill and Badoni (2003)
Salmonella	IM	Carcass	Artificial	80	10	After slaughter and chilling	1.7-2.8 cm^{-2}	Smith and Graham (1978)
	SP	Brisket	Artificial	80	10 20	After slaughter	3.4 cm^{-2} 3.6 cm^{-2}	Smith (1992)
	IM	Trimmings	Artificial	95	3	Processing facilities (before grinding)	0.7 g^{-1}	Ellebracht et al. (1999)
	SP	Carcass surface regions	Artificial	95	5	After slaughter	3.8 cm^{-2}	Castillo et al. (1998)
	SP	Beef surfaces	Artificial	72	15	After exsanguination	2.8 cm^{-2}	Cutter and Rivera-Betancourt (2000)
	IM	Trimmings[b]	Artificial	82	180	NA[c]	0.1 g^{-1}	Stivarius et al. (2002)
	IM	Muscle	Artificial	82	15	Retail	0.5 g^{-1}	Özdemir et al. (2006b)
Staphylococcus aureus	IM	Muscle	Artificial	93	15	Retail	2.5 g^{-1}	Özdemir et al. (2006a)
Yersinia enterocolitica	SP	Brisket	Artificial	80	10 20	After slaughter	3.6 cm^{-2} 3.7 cm^{-2}	Smith (1992)

[a] IM, immersion; SP, spraying.
[b] Beef ground after pasteurization with hot water.
[c] NA, not available.

Table 2: Efficacy of hot potable water in reducing bacteria counts on the surface of pork carcasses and parts

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (sec)	Sampling point	Reduction (log CFU)	Reference
Aerobic bacteria (mesophilic)	SP	Carcass side	Natural	85	10	After slaughter	1.70 cm^{-2}	Gill et al. (1998)
	SP	Carcass	Natural	85	10	After slaughter	1.38 cm^{-2}	Gill et al. (1998)
	SP	Carcass	Artificial	55	90	After slaughter	0.4 cm^{-2}	Van Netten et al. (1997)
	SP	Forequarter	Natural	90	120	After slaughter	2.32 g^{-1}	Latha et al. (2009)
	SP	Carcass	Artificial	55	90	After slaughter	0.3 cm^{-2}	Van Netten et al. (1997)
Coliforms	SP	Carcass side	Natural	85	10	After slaughter	>0.71 cm^{-2}	Gill et al. (1998)
	SP	LPT	Artificial	65.5	15	After slaughter	1.28 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	65.5	30	After slaughter	1.45 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	65.5	45	After slaughter	1.63 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	71	15	After slaughter	1.41 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	71	30	After slaughter	1.75 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	71	45	After slaughter	1.81 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	76.6	15	After slaughter	1.56 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	76.6	30	After slaughter	1.88 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	76.6	45	After slaughter	2.04 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	82.2	15	After slaughter	1.68 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	82.2	30	After slaughter	1.88 cm^{-2}	Castelo et al. (2001)
	SP	LPT	Artificial	82.2	45	After slaughter	2.29 cm^{-2}	Castelo et al. (2001)
Enterobacteriaceae	SP	Carcass	Artificial	55	90	After slaughter	0.5 cm^{-2}	Van Netten et al. (1997)
	SP	Front leg	Artificial	82.2	5	After slaughter	2 cm^{-2}	Eggenberger-Solorzano et al. (2002)

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (sec)	Sampling point	Reduction (log CFU)	Reference
Gram-negative bacteria	SP	Carcass	Artificial	55	90	After slaughter	0.3 cm^{-2}	Van Netten et al. (1997)
Gram-positive bacteria	SP	Carcass	Artificial	55	90	After slaughter	0.4 cm^{-2}	Van Netten et al. (1997)
***Lactobacillus* spp.**	SP	Carcass	Artificial	55	90	After slaughter	0.5 cm^{-2}	Van Netten et al. (1997)
Proteus vulgaricus	SP	Forequarter	Natural	90	120	After slaughter	>3.10 g^{-1}	Latha et al. (2009)
Pseudomonas aureginosa	SP	Forequarter	Natural	90	120	After slaughter	>3.13 g^{-1}	Latha et al. (2009)
Spoilage bacteria [b]	SP	Carcass [c]	Natural	60	40	After slaughter	0.10-0.98 cm^{-2}	Gill et al., 1995
	SP	Carcass [c]	Natural	75	40	After slaughter	0.86-1.94 cm^{-2}	Gill et al., 1995
	SP	Carcass [c]	Natural	80	40	After slaughter	1.37-2.19 cm^{-2}	Gill et al., 1995
	SP	Carcass [c]	Natural	85	40	After slaughter	1.63-2.90 cm^{-2}	Gill et al., 1995
	SP	Carcass [c]	Natural	90	20	After slaughter	2.19-2.90 cm^{-2}	Gill et al., 1995
	SP	Carcass [c]	Natural	90	40	After slaughter	2.05-3.30 cm^{-2}	Gill et al., 1995
	SP	Carcass [c]	Natural	90	60	After slaughter	2.20-3.30 cm^{-2}	Gill et al., 1995
	SP	Carcass [c]	Natural	90	90	After slaughter	2.12-3.22 cm^{-2}	Gill et al., 1995
Staphylococcus aureus	SP	Forequarter	Natural	90	120	After slaughter	>2.68 g^{-1}	Latha et al. (2009)
Yeasts	SP	Carcass	Artificial	55	90	After slaughter	0.2 cm^{-2}	Van Netten et al. (1997)

[a] IM, immersion; SP. Spraying; IC, immersion chilling; SC, spray chilling.
[b] Spoilage bacteria: acinetobacteria, morazellae, pseudomonads, lactobacilli and *Brochothrix thermosphacta*.
[c] Back, waste, belly, foreleg.
[d] LPT, lean pork trim.

Table 3: Efficacy of hot potable water in reducing bacteria counts on the surface of sheep/lamb carcasses and parts

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (s)	Sampling point	Reduction (log CFU)	Reference
Aerobic bacteria	SP	Carcass	Natural	54.4 82.2	10	After slaughter	0.1 cm^{-2} 1.8 cm^{-2}	Dorsa et al. (1996)
	SP	Carcass	Artificial (4 log CFU/cm^2)	54.4 82.2	10	After slaughter	1.0 cm^{-2} 1.8 cm^{-2}	Dorsa et al. (1996)
	SP	Carcass	Artificial (6 log CFU/cm^2)	54.4 82.2	10	After slaughter	2.2 cm^{-2} 3.3 cm^{-2}	Dorsa et al. (1996)
		Breast surface tissue	Artificial	74	18	After slaughter	2.7-3.2 cm^{-2}	Kochevar et al. (1997)
	SP	Carcass	Natural	83	18	Dressing	1.5 cm^{-2}	Gill et al. (1998)
	IM	Carcass	Natural	90	8	After slaughter	1.1 cm^{-2}	James et al. (2000)
Coliforms	IM	Carcass	Natural	80	10	After slaughter	1.9 cm^{-2}	Smith and Graham (1978)
	SP	Carcass	Natural	83	18	Dressing	>1.0 (10 cm)$^{-2}$	Gill et al. (1998)
Escherichia coli	IM	Mutton fleecings	Artificial	80	10	After slaughter	2.0-3.0 cm^{-2}	Smith and Graham (1978)
		Carcass	Artificial	80	10	After dressing	2.0-3.0 cm^{-2}	Smith and Graham (1978)
	SP	Carcass	Natural	83	18	Dressing	2.0 (10 cm)$^{-2}$	Gill et al. (1998)
Salmonella	IM	Mutton fleecings	Artificial	80	10	After slaughter	2.0-3.0 cm^{-2}	Smith and Graham (1978)

[a] IM, immersion; SP, spraying.

Table 4: Efficacy of hot potable water in reducing bacteria counts on the surface of poultry carcasses and parts (Revised from Loretz et al., 2010).

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (min)	Sampling point	Reduction (log CFU)	Reference
Aerobic bacteria	SP	Carcass	Natural	73	0.3	During slaughter	0.4 ml-1	Berrang et al. (2000)
	IM	Carcass	Natural	60	0.5	During slaughter	0.3–0.5 ml-1	Berrang et al. (2000)
	IM	Carcass	Natural	97	3	At retail	1.9	Avens et al. (1999)
	IM	Carcass	Natural	97	4	At retail	1.8	Avens et al. (1999)
	IM	Carcass	Natural	97	5	At retail	2.1	Avens et al. (1999)
	IM	Carcass	Natural	97	6	At retail	2.0	Avens et al. (1999)
	IM	Carcass	Natural	98	8	At retail	>3.3 cm-2	Avens et al. (2002)
	IM	Carcass	Natural	95	3	At retail	>2.8 cm-2	Avens et al. (2002)
	IM	Leg	Artificial	75	0.2	At retail	0.2 g^{-1}	Whyte et al. (2003)
	IM	Leg	Artificial	80	0.2	At retail	0.4 g^{-1}	Whyte et al. (2003)
	IM	Leg	Artificial	85	0.2	At retail	0.3 g^{-1}	Whyte et al. (2003)
	IM	Leg	Artificial	70	0.3	At retail	0.6 g^{-1}	Whyte et al. (2003)
	IM	Leg	Artificial	80	0.3	At retail	0.7 g^{-1}	Whyte et al. (2003)
	IM	Leg	Artificial	85	0.3	At retail	1.1 g^{-1}	Whyte et al. (2003)
	IM/SC	Carcass	Natural	70/12–15	0.7/0.2	After slaughter	1.1–1.3 ml^{-1}	Purnell et al. (2004)
	IM	Carcass	Natural	80 or 65	0.3 or 1.2	After slaughter	0.9 ml^{-1}	Purnell et al. (2004)
	IM/SC	Carcass	Natural	75/12–15	0.5/0.2	After slaughter	0.7 ml^{-1}	Purnell et al. (2004)
	IM	Carcass	Natural	65–75	0.5	After slaughter	0.6–1.1 ml^{-1}	Purnell et al. (2004)

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (min)	Sampling point	Reduction (log CFU)	Reference
	IM	Carcass	Natural	70	1	During slaughter	1.3 cm^{-2}	Sinhamahapatra et al. (2004)
	SP	Carcass	Natural	70	1	During slaughter	1.2 cm^{-2}	Sinhamahapatra et al. (2004)
	SP	Carcass	Artificial	21–54	0.1	During slaughter	2.1–2.3 ml^{-1}	Northcutt et al. (2005)
	IM	Carcass	Natural	95	0.5–4	At retail	1.2–3.1 cm^{-2}	Tompkins et al. (2008)
***Campylobacter* spp.**	IM	Carcass	Natural	60	0.5	During slaughter	<0.5 ml^{-1}	Berrang et al. (2000)
	SP	Carcass	Natural	70–73	0.3	During slaughter	0.1–0.4 ml^{-1}	Berrang et al. (2000)
	IM/SC	Carcass	Natural	70/12–15	0.7/0.2	After slaughter	1.1–1.6 ml^{-1}	Purnell et al. (2004)
	IM	Carcass	Natural	75–80	0.3–0.5	After slaughter	0.9–1.1 ml^{-1}	Purnell et al. (2004)
	SP	Carcass	Artificial	21–54	0.1	During slaughter	2.1–2.8 ml^{-1}	Northcutt et al. (2005)
Campylobacter jejuni	SP/IC	Carcass	Artificial	55–60	0.2/50	During slaughter	2.4–2.5/carcass	Li et al. (2002)
	SP	Carcass	Artificial	55–65	0.2–0.3	During slaughter	1.3–1.4/carcass	Li et al. (2002)
	IM	Carcass	Artificial	55–65	0.3	During slaughter	0.6–1.4 cm^{-2}	Li et al. (2002)
	IM	Leg	Artificial	75	0.2	At retail	0.92 g^{-1}	Whyte et al. (2003)
	IM	Leg	Artificial	75	0.3	At retail	0.87 g^{-1}	Whyte et al. (2003)
	IM	Leg	Artificial	80	0.2	At retail	0.84 g^{-1}	Whyte et al. (2003)
	IM	Leg	Artificial	80	0.3	At retail	1.77 g^{-1}	Whyte et al. (2003)
	IM	Leg	Artificial	85	0.2	At retail	1.08 g^{-1}	Whyte et al. (2003)
	IM	Leg	Artificial	85	0.3	At retail	1.89 g^{-1}	Whyte et al. (2003)
	IM	Carcass	Artificial	75	0.5	After chilling	1.7 cm^{-2}	Corry et al. (2007)

Microorganism	Application[a]	Treated material	Contamination	Temperature (°C)	Exposure time (min)	Sampling point	Reduction (log CFU)	Reference
	IM	Carcass	Artificial	80	0.3	After chilling	1.3 cm^{-2}	Corry et al. (2007)
	IM	Carcass	Artificial	70	0.7	After chilling	1.0 cm^{-2}	Corry et al. (2007)
		Carcass	Artificial	80	0.3	After chilling	1.63–2.91 cm^{-2}	James et al. (2007)
Coliforms	IM	Carcass	Natural	60–70	0.3–0.5	During slaughter	0.3–0.5 ml^{-1}	Berrang et al. (2000)
	SP	Carcass	Natural	73	0.3	During slaughter	0.1 ml^{-1}	Berrang et al. (2000)
	IM	Carcass	Natural	70	1	During slaughter	1.3 cm^{-2}	Sinhamahapatra et al. (2004)
	SP	Carcass	Natural	70	1	During slaughter	0.7 cm^{-2}	Sinhamahapatra et al. (2004)
Enterobacteriaceae	IM	Leg	Artificial	75–85	0.2–0.3	At retail	0.1–0.7 g^{-1}	Whyte et al. (2003)
	IM/SC	Carcass	Natural	75/12–15	0.5/0.2	After slaughter	>1.1 ml^{-1}	Purnell et al. (2004)
	IM	Carcass	Natural	70–80	0.3–0.7	After slaughter	0.7–1.0 ml^{-1}	Purnell et al. (2004)
	IM	Carcass	Natural	65	0.5–1.2	After slaughter	0.5–0.7 ml^{-1}	Purnell et al. (2004)
	IM/SC	Carcass	Natural	70/12–15	0.7/0.2	After slaughter	0.2–1.6 ml^{-1}	Purnell et al. (2004)
Escherichia coli	IM	Carcass	Natural	60–70	0.3–0.5	During slaughter	0.5–0.7 ml^{-1}	Berrang et al. (2000)
	SP	Carcass	Natural	73	0.3	During slaughter	0.1 ml^{-1}	Berrang et al. (2000)
	SP	Carcass	Artificial	21–54	0.1	During slaughter	1.8–2.1 ml^{-1}	Northcutt et al. (2005)
	IM	Carcass	Artificial	70–80	0.3–0.7	After chilling	1.2–1.3 cm^{-2}	Corry et al. (2007)
		Carcass	Artificial	80	0.3	After chilling	1.63–2.95 cm^{-2}	James et al. (2007)
***Salmonella* spp.**	SP	Carcass	Artificial	21–54	0.1	During slaughter	0.7–1.2 ml^{-1}	Northcutt et al. (2005)

[a] IM, immersion; SP, spraying; IC, immersion chilling; SC, spray chilling.

Table 5: Efficacy of recycled hot water in reducing bacteria counts on the surface of pork and beef carcasses

Microorganism	Treated material	Application[a]	Sampling point	Contamination	Temperature (°C)	Exposure time (sec)	Reduction (log CFU)	Reference
				Pork Carcasses				
Aerobic bacteria	Carcass (except anal area site)	IM	After slaughter	Natural	85	15	1.78-1.94 cm^{-2}	Gill et al. (1997)
	Carcass (anal area site)	IM	After slaughter	Natural	85	15	0.09-0.65 cm^{-2}	Gill et al. (1997)
Coliforms	Carcass (anal area site)	IM	After slaughter	Natural	85	15	1.36-1.74 cm^{-2}	Gill et al. (1997)
Escherichia coli	Carcass (anal area site)	IM	After slaughter	Natural	85	15	0.69-1.33 cm^{-2}	Gill et al., (1997)
				Beef Carcasses				
Aerobic bacteria	Various sites	IM	After slaughter	Natural	85	8-12	0.82-1.56 cm^2	Gill and Bryant (2000)
Coliforms	Various sites	IM	After slaughter	Natural	85	8-12	0.98-2.33 cm^2	Gill and Bryant (2000)
Escherichia coli	Various sites	IM	After slaughter	Natural	85	8-12	1.1-2.8 cm^2	Gill and Bryant (2000)

Figure 1: Effect of water temperature and exposure time on the reduction of coliform numbers after hot water (HW) decontamination of artificially contaminated pork based on data of Castelo et al. (2001)

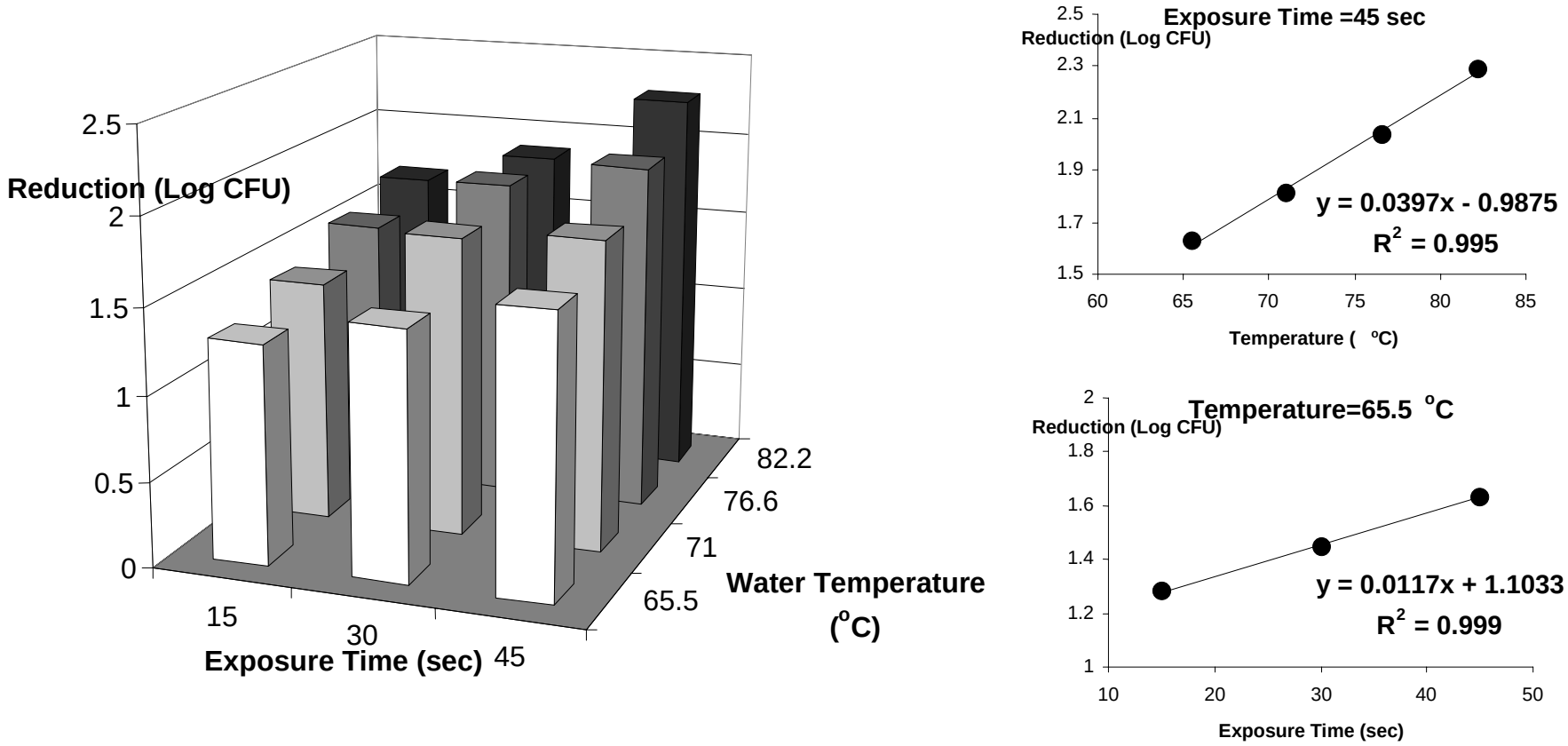


Figure 2: (a) Operating characteristic curve of the EU Process Hygiene Criteria (PHC) for Total Viable count (TVC) in Steer/Heifer/Cow/Bull/Calf (n=5, m=3.0 log CFU/cm^2, M=5.0 log CFU/cm^2) carcasses. (b) Effect of hot water (HW) decontamination on the compliance with the PHC for carcasses with mean TVC concentration of 6.0 log CFU/cm^2

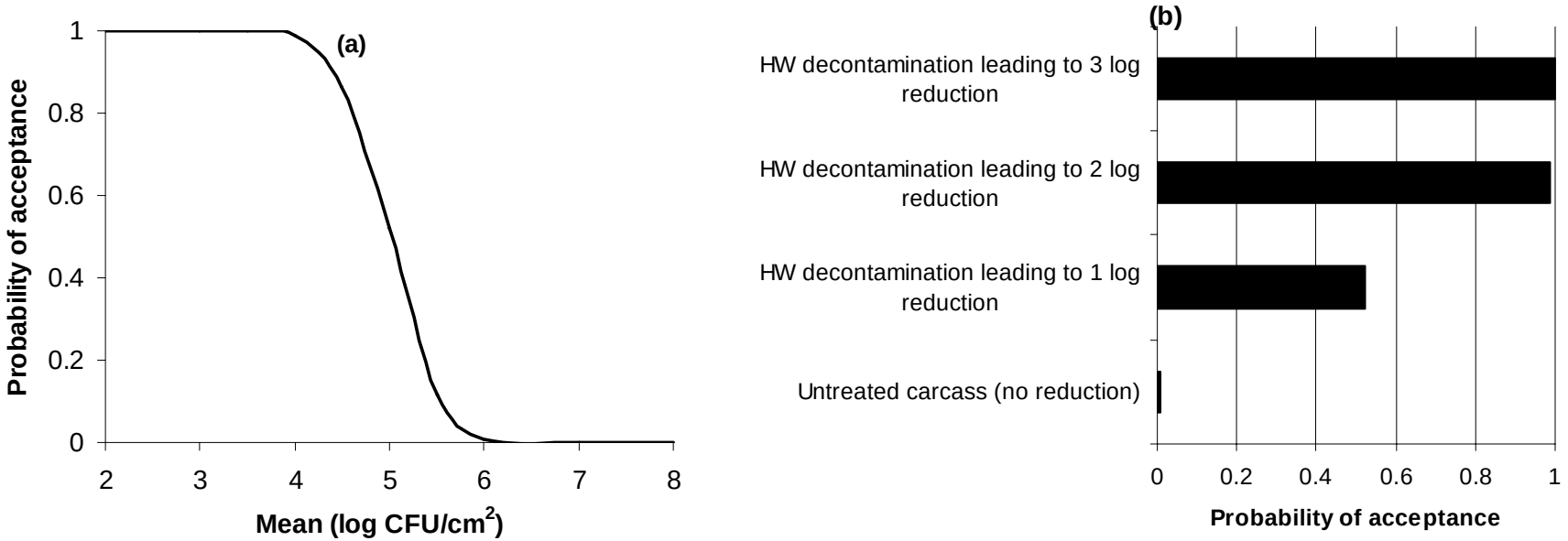
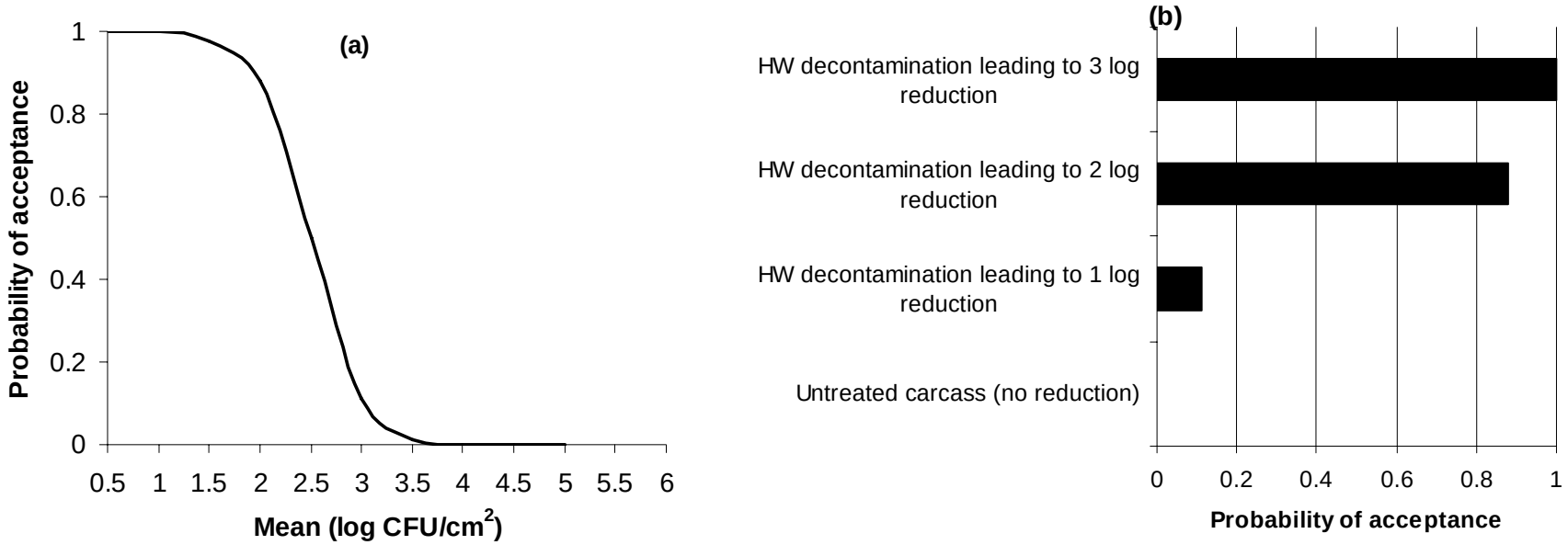


Figure 3: (a) Operating characteristic curve of the EU Process Hygiene Criteria (PHC) for Enterobacteriaceae in Steer/Heifer/Cow/Bull (n=5, m=1.5 log CFU/cm2, M=2.5 log CFU/cm2) carcasses. (b) Effect of hot water (HW) decontamination on the compliance with the PHC for carcasses with mean TVC concentration of 4 log CFU/cm2



GLOSSARY

- **Abiotic risk**: Risk derived by abiotic components which are, in biology, non-living chemical and physical factors in the environment.

- **Efficacy:** for the purpose of this opinion the efficacy of the hot water decontamination treatment is defined as the extent of reduction of microbial counts achieved on the carcass surface.

- **Contaminants** are substances present in food which, in sufficient concentration, can adversely affect living organisms and are not intentionally added to such food as a result of the production (including operations carried out in crop husbandry, animal husbandry and veterinary medicine), manufacture, processing, preparation, treatment, packing, packaging, transport or holding of such food, or as a result of environmental contamination. Extraneous matters, such as, for example, insect fragments, animal hair, etc, are not covered by this definition.

- **Decontamination treatment**: process applied to remove or reduce surface contamination of food.

- **Deluge cabinets** a cabinet involving a deluge, or waterfall, method of hot water distribution on meat carcasses.

- **HACCP (Hazard Analysis Critical Control Point)** is a systematic preventive approach to food safety and pharmaceutical safety that addresses physical, chemical, and biological hazards as a means of prevention rather than finished product inspection. HACCP is used in the food industry to identify potential food safety hazards, so that key actions, known as Critical Control Points (CCPs) can be taken to reduce or eliminate the risk of the hazards being realized. The HACCP principles are:

    - **Hazard analysis** plans determine the food safety hazards and identify the preventive measures the plan can apply to control these hazards. A food safety hazard is any biological, chemical, or physical property that may cause a food to be unsafe for human consumption.

    - **Critical control points** A Critical Control Point (CCP) is a point, step, or procedure in a food manufacturing process at which control can be applied and, as a result, a food safety hazard can be prevented, eliminated, or reduced to an acceptable level.

    - **Critical limits** is the maximum or minimum value to which a physical, biological, or chemical hazard must be controlled at a critical control point to prevent, eliminate, or reduce to an acceptable level.

    - **Monitoring activities** are necessary to ensure that the process is under control at each critical control point. Each monitoring procedure and its frequency should be listed in the HACCP plan.

    - **Corrective action**s are to be taken when monitoring indicates a deviation from an established critical limit. The final rule requires a plant's HACCP plan to identify the corrective actions to be taken if a critical limit is not met. Corrective actions are intended to ensure that no product injurious to health or otherwise adulterated as a result of the deviation enters commerce.

    - **Record keeping:** The HACCP regulation requires that all plants maintain certain documents, including its hazard analysis and written HACCP plan, and records documenting the monitoring of critical control points, critical limits, verification activities, and the handling of processing deviations.

- **Verification** ensures the HACCP plan is adequate, and working as intended. Verification procedures may include such activities as review of HACCP plans, CCP records, critical limits and microbial sampling and analysis. Verification also includes 'validation' - the process of finding evidence for the accuracy of the HACCP system (e.g. scientific evidence for critical limitations).

- **Validation** ensures that the plants do what they were designed to do; that is, they are successful in ensuring the production of safe product. Plants will be required to validate their own HACCP plans. FSIS will not approve HACCP plans in advance, but will review them for conformance with the final rule.

- **MRLs (Maximum Residual Level):** upper legal levels of a concentration for pesticide or drugs residues in or on food or feed. MRLs are set for a wide range of food commodities of plant and animal origin, and they usually apply to the product as put on the market. MRLs are not simply set as toxicological threshold levels, but they are derived after comprehensive assessment of the properties of the active substance and the residue behaviour on treated crops. An indispensable precondition for setting MRLs is the performance of a risk assessment to ensure consumer safety.

- **Potable water** is referred to water meeting the minimum requirements laid down in Council Directive 98/83/EC of 3 November 1998 on the quality of water intended for human consumption. **Clean water** means clean seawater and freshwater of a similar quality.

- **Spore** is a biological structure that is adapted for dispersal and surviving for extended periods of time in unfavourable conditions. Spores form part of the life cycles of many bacteria, plants, algae, fungi and some protozoans. Concerning bacterial spores, the endospore is a dormancy form that the bacterium can reduce itself to, when the environment becomes deleterious for the bacterial vegetative state, notably including dessication. Examples of spore-forming bacteria of public health importance are the genus *Clostridium* spp. and *Bacillus* spp.

- **Tyndallisation**: method developed to destroy bacteria and spores by exposure to two or three sequential heating treatments lower than those needed for direct inactivation of spores by sterilization at temperatures above 100°C.

- **Veterinary medicinal products**[15] any substance or combination of substances presented for treating or preventing disease in animals; any substance or combination of substances which may be administered to animals with a view to making a medical diagnosis or to restoring correcting or modifying physiological functions in animals is likewise considered a veterinary medicinal product.

15 Directive 2001/82/EC of the EP and of the Co of 6 November 2001 on the Community code relating to veterinary medicinal product